

CHAPTER 12

LABORATORY

STANDARD OPERATING PROCEDURE

500 BED FLEET HOSPITAL

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500 BED FLEET HOSPITAL
STANDARD OPERATING PROCEDURES OUTLINE
LABORATORY DEPARTMENT

A. **MISSION:** Provide a broad spectrum of diagnostic testing for the treatment of both medical and surgical patients.

B. **FUNCTIONS:**

1. Coordinate all laboratory functions within the Fleet Hospital.
2. Perform laboratory procedures related to hematology, microbiology, chemistry, and urinalysis.
3. Perform routine blood typing (ABO & RH), routine crossmatches, limited blood drawing and processing, blood storage, frozen plasma storage.

C. **PHYSICAL DESCRIPTION:**

1. Head, Laboratory Department.
 - (a) Location within complex:
 - (b) Sheltering.
Type: Temper Tent.
Quantity:
 - (c) Material.
IOL:
2. Laboratory Module.
 - (a) Location within complex:
 - (b) Sheltering.
Type: Expandable, Hardwall Shelter.
Quantity:
 - (c) Material.
IOL:
3. Blood Bank.
 - (a) Location within complex:
 - (b) Sheltering.
Type: Expandable, Hardwall Shelter.
Quantity:
 - (c) Material.

IOL:

4. Laboratory Support Space.

(a) Location within complex:

(b) Sheltering.

Type: Temper Tent.

Quantity:

(c) Material.

IOL:

5. Morgue.

(a) Location within complex:

(b) Sheltering.

Type: Tent of opportunity or refrigerated van as required.

Quantity:

(c) Material.

IOL:

D. **SPECIAL CONSIDERATIONS:**

1. No anatomical pathology capability.

2. No permanent morgue facility.

3. CBC will include HCT, HGB, and WBC only.

4. Sed rates and platelet counts will not be performed. Platelet estimates will be reported from the differential slide when ordered.

5. No chemistry panels will be performed, only individual tests will be ordered and will be subject to review by a pathologist (i.e., NA, K, CL, or CO2 instead of electrolytes).

6. Buns will only be performed on patients with renal problems or failure (12) or every other day on surgical patients (64).

7. Urinalysis's will only be performed on patients upon admission (44), for urinary tract infections, renal failure, or every other day on surgical patients (106).

8. Creatinine's will only be performed on patients with renal failure or with urinary or renal difficulties.

9. Amylase's will only be performed in cases of abdomen trauma, pancreatitis or every other day after major abdomen surgery.

10. Due to the very limited supply of fresh frozen plasma, it will only be available for patients with bleeding difficulties. Albumin or other blood volume expanders will be used as a replacement.

11. The lab will have limited blood drawing and specimen collection capabilities. All samples for testing will be collected and delivered to the lab by each department (i.e., ward, I.C.U., clinic, and casualty receiving).

12. Blood and fresh frozen plasma will be supplied to the Blood Bank through the DOD Blood Program. The Blood Bank will have very limited blood drawing capabilities (72 units).

13. All crossmatched blood will be picked up from the Blood Bank by the department ordering it.

E. **WORKLOAD:**

1. 120 crossmatches.
2. Draw 72 units of blood.
3. Store 600 units of blood.
4. Store 100 units of plasma.
5. Average test frequencies: (Peak loads are 1/3 higher.)

Test

CBC
Diff
PT/PTT
Glucose
Bun
Electrolytes
UA
Cultures
Sensitivity
Creatinine
Amylase
SGOT
SGPT
Monospot
Blood Culture
CO₂
Platelet Estimate

F. **ORGANIZATION:**

1. Responsibility. The Head, Laboratory Department, who reports to the Director of Ancillary Services, is assigned overall management responsibility. The department is divided into two divisions.

2. Organizational chart.

COMMANDING OFFICER

DIRECTOR, ANCILLARY SERVICES

HEAD, LABORATORY DEPARTMENT/
LABORATORY OFFICER

WATCH LPO

MODULE 1 LPO

RECEIVING
TECHNICIAN

MODULE 2 LPO

CHEMISTRY MICROBIOLOGY/
URINALYSISBLOOD
BANKHEMATOLOGY/
SEROLOGY

3. Staffing.

(a) Criteria.

(1) Two 12 hour shifts, six days a week. Sundays covered by a duty crew.

(2) Peak workload assumed to occur on AM shift.

(3) One officer assigned to each shift.

(4) Morgue functions covered on an on-call basis, as needed.

(5) Personnel must be capable of working in every section of the lab.

(6) Every four weeks, crews completely switch shifts.

(b) Staffing pattern. Two 12 hour watches.

<u>PERSONNEL</u>	<u>AM</u>	<u>NW</u>	<u>BILLET #</u>	<u>TOTAL</u>
Head, Laboratory Dept	1	-	70020	1
Laboratory Officer	1	1	72020	2
Watch LPO	1	1	72010, 72030.00	2
LAB 1 & 2 LPO's	2	2	72030.01-.04	4
Adv. Lab Tech (E-6)	3	2		5
Adv. Lab Tech (E-5)	8	8		16
Adv. Lab Tech (E-4)	.4	4		8
Basic Lab Tech (E-4)	4	3		7

4. Assignments by billet sequence number: See TAB A, page 14.

5. Watch bill: See TAB B, page 15.

6. Special watches: N/A.

G. TASKS:

TASK	METHOD
1. ACCEPT SPECIMEN DELIVERED TO THE LABORATORY	1.1 Give priority to STAT or ASAP laboratory requests.
	1.2 Verify acceptability of specimen IAW TAB C-1.
	1.3 Place routine specimens and lab slips in the appropriate rack IAW TAB C-2
	1.4 Walk STATS to the section of the lab performing that test.

- | | | |
|--|-------|---|
| | 1.5 | Direct runner to walk blood bank and blood gas specimens directly to those sections of the lab. |
| 2. ACCESSION SPECIMEN
(EXCEPT BLOOD BANK SPECIMENS) | 2.1 | Record the next sequential number from the accession log on the: |
| | 2.1.A | Request slip. |
| | 2.1.B | Specimen container. |
| | 2.1.C | Accession log |
| | 2.2 | Record patient/test request information in the accession log IAW TAB C-3. |
| 3. RECORD RESULTS | 3.1 | Record test results legibly onto: |
| | 3.1.A | Request slip. |
| | 3.1.B | Accession log IAW TAB C-3. |
| 4. REPORT RESULTS | 4.1 | Report all critical values by telephone (see TAB C-4). |
| | 4.2 | Report all STATS by telephone. |
| | 4.3 | File lab slips for pickup by patient care personnel (see TAB C-5). |
| 5. PERFORM HEMATOLOGY PROCEDURES | 5.1 | Perform CBC IAW TABS C-6 through C-9. |
| | 5.2 | Perform differential and platelet estimate IAW TABS C-10 - C-12 when indicated. |
| | 5.3 | Perform body fluid cell counts IAW TABS C-13-14. |
| | 5.4 | Perform screen for malaria IAW TAB C-15. |
| 6. PERFORM COAGULATION PROCEDURES | 6.1 | Perform prothrombin time (PT) IAW TABS C-16 and C-17. |
| | 6.2 | Perform partial thromboplastin time (PTT) IAW TABS C-16 and C-18. |
| | 6.3 | Perform fibrinogen determination IAW TAB C-19. |
| 7. PERFORM URINALYSIS PROCEDURES | 7.1 | Perform complete urinalysis procedures IAW TAB C-20. |
| | 7.2 | Perform urine pregnancy test IAW |

		TAB C-21.
8. PERFORM SEROLOGY PROCEDURES TAB C-22.	8.1	Perform mononucleosis test IAW
	8.2	Perform RPR test IAW TAB C-23.
9. PERFORM MICROBIOLOGY 9.1 PROCEDURES		Plant culture according to specimen source IAW TAB C-24.
	9.2	Identify organism IAW TABS C-25 through C-35.
	9.3	Perform gram stains as required IAW TAB C-25.
	9.4	Perform bacterial antibiotic sensitivity testing IAW TABS C-36 and C-37.
	9.5	Perform blood culture procedures IAW TAB C-24.
	9.6	Perform parasitology and fungal procedures IAW TABS C-24 and C- 25.
10. PERFORM CHEMISTRY PROCEDURES	10.1	Determine sodium / potassium (NA+/K+) levels IAW TAB C-31.
	10.2	Determine chloride level IAW TAB C-32.
	10.3	Determine carbon dioxide level IAW TAB C-33.
	10.4	Determine other chemistry levels (glucose, BUN, Creatinine, Amylase, SGOT, SGPT) IAW TABS C- 34 through C-39.
11. PERFORM ROUTINE BLOOD BANK PROCEDURES	11.1	Accession Blood Bank specimens IAW TABS C-47, F-2 and F-10.
	11.2	Perform blood grouping and typing IAW TABS C-40 through C- 46, F-2 and F-10.
	11.3	Perform crossmatch procedure IAW TABs C-46, F-2 and F-10.
	11.4	Collect, process and store blood IAW TABs C-50 - C-55, F-2 and F- 10.
	11.5	Issue blood for transfusion IAW TABs C-56, F-2 and F-10.
	11.6	Receive incoming blood shipments IAW TAB C-55.
	11.7	Receive returned blood bags from ward IAW TABS C-55, F-2 and F-

		10.
	11.8	Perform transfusion reaction workup IAW TABs C-56, F-2 and F-10.
	11.9	Communication with Theater Blood Coordinator IAW TAB C-56.
12. RECEIVE MORGUE ADMISSIONS	12.1	Receive morgue admissions IAW TAB C-57.
13. SHIP MAILOUT SPECIMENSshipment.	13.1	Package histology or other Prepare required request form or consult form.
	13.2	Initiate appropriate communications regarding shipment.
	13.3	Ship IAW Chapter 10.
14. COLLECT SPECIMENS FROM AMBULATORY PATIENTS	14.1	Verify completeness of request as described in TAB C-1.
	14.2	Verify identify of individual.
	14.3	Draw blood IAW TABs C-58 and C-59.
	14.4	Collect urine sample IAW TAB C-60.
15. PERFORM LABORATORY ADMINISTRATIVE FUNCTIONS	15.1	Provide personnel.
	15.1.A	Determine staffing needs and post schedule to assure present-in-section or on-call coverage for service at all times.
	15.1.B	Recall staff IAW TAB C-61.
	15.1.C	Provide training as necessary to assure that duty personnel have the required skills to accomplish the mission.
	15.2	Maintain a reference library, available to hospital staff as well as laboratory personnel.
	15.2.A	As a minimum, maintain all references listed in TAB F.
	15.2.B	Update and maintain all department procedure manuals.
	15.3	Maintain general files IAW TAB C-63.
16. MAINTAIN WORKING	16.1	Identified working levels

LEVELS OF SUPPLIES/EQUIPMENT		of supplies.
	16.1.A	Accomplish request/requisitions/return functions IAW with Chapter 10.
	16.1.B	Ensure that supplies on hand do not exceed identified levels under normal circumstances.
	16.1.C	File copies of supply documents.
	16.2	Store supplies properly, IAW manufacturer's instructions.
	16.2.A	Maintain equipment accountability at all times.
17. PREPARE REPORTS	17.1	Prepare Blood Bank operational report IAW TAB C-65.
18. KEEP LAB SPACES CLEAN	18.1	Dispose of contaminated waste IAW TAB C-66.
	18.2	Dispose of needles IAW TABs C-71 and C-72.
	18.3	Decontaminate lab counters daily.
	18.4	Field day the laboratory.
19. PERFORM EQUIPMENT MAINTENANCE	19.1	Perform equipment maintenance for all equipment IAW manufacturer's instructions.
	19.2	Report maintenance requirements not specified as operator maintenance to general or medical maintenance personnel.
	19.3	Maintain appropriate records.
20. MAINTAIN DEPARTMENTAL LOG	20.1	The LPO of the watch will maintain the departmental log. He will:
	20.1.A	Document significant events such as: <ul style="list-style-type: none"> - Fire. - Personal injury. - Staff musters. - Utility failures. - Significant equipment failures. - Field day activities. - Watch reliefs. - Recalls. - Medical emergencies. - Crash cart inspections. - Safety deficiencies. - Other appropriate events.

- | | | | |
|-----|-----------------------------------|--------|--|
| 21. | PREPARE STAINS AND REAGENT | 21.1 | Prepare stains and reagents following manufacturer's specifications. |
| 22. | PREPARE BACTERIOLOGY MEDIA | 22.1 | Prepare bacteriology media following manufacturer's specifications and IAW TABs C-24 - C-29. |
| 23. | PERFORM QUALITY CONTROL FUNCTIONS | 23.1 | Record temperatures of all refrigerators, freezers, incubators and water baths IAW TAB C-70. |
| | | 23.2 | Run commercial/patient control IAW TAB C-67. |
| | | 23.3 | Review all QC data periodically IAW TAB C-67. |
| | | 23.4 | Test spore strips from CSR IAW TAB C-30. |
| | | 23.5 | Calibrate machines following manufacturer's recommendations. |
| 24. | REACT TO MEDICAL EMERGENCIES | 24.1 | Personnel must recognize medical emergencies, to include shock, hemorrhage, pulmonary or cardiopulmonary arrest, partial airway obstruction, adverse reaction to blood collecting and simple fainting. |
| | | 24.1.A | Treat these IAW TAB C-62. |
| | | 24.2 | All personnel must be able to locate emergency equipment and emergency tray immediately and initiate oxygen therapy. Obtain help as needed to manage the emergency. |
| 25. | MANAGE LAB REPORTS | 25.1 | File lab copies of request slips IAW TAB C-5. |
| | | 25.2 | Maintain lab logs IAW TAB C-5. |
- H. **STANDARD OPERATING PROCEDURES:** See TAB C, page 16.
- I. **CLINICAL POLICIES/GUIDELINES:** See TAB D, page 191.
- J. **STANDARDS AND JOB DESCRIPTIONS:** See TAB E, page 194.
- K. **DOCUMENTATION:**
1. References: See TAB F, page 207.
 2. Forms: See TAB G, page 208.

TAB A
ASSIGNMENTS BY BILLET SEQUENCE CODE

Department: LABORATORY

<u>Billet Number</u>	<u>Title</u>	<u>Designator</u>	<u>Rank/ Rate</u>	<u>Watch Section</u>
70029	HEAD, LABORATORY DEPT	2100	0-5	WATCHBILL
72049	MEDICAL TECHNOLOGIST	2300	0-4	WATCHBILL
72069	MEDICAL TECHNOLOGIST	2300	0-3	WATCHBILL
72019	ADVANCED LAB TECH	8506/HM	E-7	WATCHBILL
72039	ADVANCED LAB TECH	8506/HM	E-6	WATCHBILL
72041	ADVANCED LAB TECH	8506/HM	E-6	1
72042	ADVANCED LAB TECH	8506/HM	E-6	3
72043	ADVANCED LAB TECH	8506/HM	E-6	5
72045	ADVANCED LAB TECH	8506/HM	E-6	7
72047	ADVANCED LAB TECH	8506/HM	E-6	2
72049	ADVANCED LAB TECH	8506/HM	E-6	4
72051	ADVANCED LAB TECH	8506/HM	E-6	6
72052	ADVANCED LAB TECH	8506/HM	E-6	8
72053	ADVANCED LAB TECH	8506/HM	E-6	1
72059	ADVANCED LAB TECH	8506/HM	E-5	2
72061	ADVANCED LAB TECH	8506/HM	E-5	4
72062	ADVANCED LAB TECH	8506/HM	E-5	1
72063	ADVANCED LAB TECH	8506/HM	E-5	4
72064	ADVANCED LAB TECH	8506/HM	E-5	1
72065	ADVANCED LAB TECH	8506/HM	E-5	6
72066	ADVANCED LAB TECH	8506/HM	E-5	8
72067	ADVANCED LAB TECH	8506/HM	E-5	8
72068	ADVANCED LAB TECH	8506/HM	E-5	5
72069	ADVANCED LAB TECH	8506/HM	E-5	7
72071	ADVANCED LAB TECH	8506/HM	E-5	2
72073	ADVANCED LAB TECH	8506/HM	E-5	3
72075	ADVANCED LAB TECH	8506/HM	E-5	5
72077	ADVANCED LAB TECH	8506/HM	E-5	6
72087	ADVANCED LAB TECH	8506/HM	E-5	7
72089	ADVANCED LAB TECH	8506/HM	E-5	3
72079	ADVANCED LAB TECH	8506/HM	E-4	3
72081	ADVANCED LAB TECH	8506/HM	E-4	3
72082	ADVANCED LAB TECH	8506/HM	E-4	7
72083	ADVANCED LAB TECH	8506/HM	E-4	6
72084	ADVANCED LAB TECH	8506/HM	E-4	8
72085	ADVANCED LAB TECH	8506/HM	E-4	1
72091	ADVANCED LAB TECH	8506/HM	E-4	2
72093	ADVANCED LAB TECH	8506/HM	E-4	4
72099	BASIC LAB TECH	8501/HM	E-4	4
72101	BASIC LAB TECH	8501/HM	E-4	2
72103	BASIC LAB TECH	8501/HM	E-4	5
72105	BASIC LAB TECH	8501/HM	E-4	6
72107	BASIC LAB TECH	8501/HM	E-4	7
72109	BASIC LAB TECH	8501/HM	E-4	8
72111	BASIC LAB TECH	8501/HM	E-4	5

TAB B
WATCH BILL FOR LABORATORY DEPARTMENT

Section	M	T	W	T	F	S	S	M	T	W	T	F	S	S	M	T	W	T	F	S	S
1	A	A	A	A	A	A	A	A	A	A	A	A	A	*	A	A	A	A	A	A	A
2	A	A	A	A	A	A	A	A	A	A	A	A	A	-	A	A	A	A	A	A	A
3	A	A	A	A	A	A	*	A	A	A	A	A	A	A	A	A	A	A	A	A	-
4	A	A	A	A	A	A	-	A	A	A	A	A	A	A	A	A	A	A	A	A	*
5	P	P	P	P	P	P	P	P	P	P	P	P	P	*	P	P	P	P	P	P	P
6	P	P	P	P	P	P	P	P	P	P	P	P	P	-	P	P	P	P	P	P	P
7	P	P	P	P	P	P	*	P	P	P	P	P	P	P	P	P	P	P	P	P	-
8	P	P	P	P	P	P	-	P	P	P	P	P	P	P	P	P	P	P	P	-	*
0-5	A	A	A	A	A	-	A	A	A	A	A	A	A	-	A	A	A	A	A	A	-
0-4	P	P	P	P	P	P	-	P	P	P	P	P	P	P	P	P	P	P	P	P	*
0-3	-	A	A	A	A	A	A	-	A	A	A	A	A	A	-	A	A	A	A	A	A
E-7	A	A	A	A	A	A	-	A	A	A	A	A	A	*	A	A	A	A	A	A	A
E-6	P	P	P	P	P	P	*	P	P	P	P	P	P	-	P	P	P	P	P	P	-

KEY:

A = 0700-1900

P = 1900-0700

* = On Call

- = Excused

TAB C
STANDARD OPERATING PROCEDURES INDEX

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C-27	Coagulase Test	75
C-28	Oxidase Test	77

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C-32	Chloride	83
C-33	CO ₂ Determination	86.
C-34	Glucose	88
C-35	BUN	90
C-36	Creatinine	92
C-37	Amylase	95
C-38	SGOT	97
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TAB C-1

UNACCEPTABLE SPECIMENS

A. **PURPOSE:** To identify conditions for which specimens will not be accepted by the laboratory for testings and to specify corrective measures to be taken in these cases.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:** N/A.

D. **CRITERIA:**

1. Specimens must be clearly labeled with:

(a) The patient's register number as the very minimum.

(b) If known, name (first and last) must also be indicated.

(c) Specimens collected for blood product compatibility testing require additional information.

2. The minimum acceptable information on the request slip includes:

(a) Patient registration number (and name if known).

(b) Patient location (ward).

(c) Requesting physician.

(d) Time and date collected.

(e) Test requested.

(f) Priority (routine, ASAP, or STAT).

3. Specimens must be collected properly for the test requested. Examples of unacceptable samples include:

(a) Anticoagulated specimens for tests requiring serum.

(b) Clotted specimens for test requiring whole blood or plasma.

(c) Insufficient blood in a tube in which the specimen/anti-coagulant ratio is critical.

(d) Insufficient specimen for the test requested.

(e) Specimens obtained from IV lines.

(f) Hemolyzed or lipemia blood specimens for tests which are affected by hemolysis or lipemia.

E. **STEPS:**

1. Do not process unacceptable specimens.

2. Notify the runner of the discrepancy.
3. Direct the patient care staff to properly recollect the specimen or to indicate the information missing from the slip.

F. **RESPONSIBILITY:**

1. The requesting physician is responsible for properly collecting and submitting specimens for testing.
2. The Head, Laboratory Department is responsible for verifying the acceptability of specimens.

TAB C-2

SPECIMEN RECEIVING RACKS

A. **PURPOSE:** To describe the method and use of racks for the receipt of specimens.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Test tube racks (2).
2. Test tube rocker (1).
3. Basins/boxes/or trays (2).

D. **CRITERIA:**

1. Specimens are separated by test/lab section upon receipt to facilitate testing.
2. Labs slips accompany specimens.

E. **STEPS:**

1. Hematology and coagulation specimens are placed onto the test tube rocker, from left to right as received.
2. Chemistry and serology specimens are placed into separate racks labeled as "chemistry" and "serology." Place them from left to right, front to back, as received.
3. Microbiology and urinalysis specimens are placed onto separate trays labeled "microbiology" and "urinalysis." Place urine specimens requiring both a routine UA and a C & S onto the urinalysis tray.
4. Place request slips in piles face down next to each rack or tray.

F. **RESPONSIBILITY:**

1. Each section will periodically pickup specimens from racks in the receiving area.
2. The technician in the receiving area is responsible to deliver STATS directly to each section and to direct the runner to deliver Blood Bank and blood gas specimens to those sections.

TAB C-3

SECTION ACCESSIONING LOGS

A. **PURPOSE:** To provide a sequential, chronological, legal record of tests performed in each section of the laboratory.

B. **DEFINITION:** A hardbound log (record book) containing the minimum essential information required to identify lab test and results.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

Standard record books (7).

D. **CRITERIA:**

1. One log will be maintained for each functional section of the laboratory, (hematology, urinalysis, chemistry, serology, microbiology, Blood Bank, blood gases, and mail-outs).

2. Logs will be maintained concurrently with the performance of tests, when possible. At a minimum, they will be updated before the watch LPO is relieved.

E. **STEPS:**

1. Mark the front cover of the log with the Fleet Hospital UIC, the title "Hematology Accession Log" (or appropriate section), and the date of initial entry.

2. Divide each set of facing pages into vertical columns. Label columns IAW TABs G-20 through G-27.

TAB C-4

CRITICAL VALUES

A. **PURPOSE:** To identify critical laboratory values and the mechanism to report such values to appropriate patient care personnel.

B. **DEFINITION:** A critical value is one which may be life threatening to a patient and therefore must be brought to the attention of a health care provider for immediate patient treatment or action.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:** N/A.

D. **CRITERIA:**

1. Critical values will be posted in each lab module for easy reference.
2. Critical values will be brought to the attention of the module LPO.
3. All critical values will be verified before being reported.
4. Appropriate personnel will be notified directly (by telephone or in-person) of all critical lab values.

5. The following laboratory values are considered critical:

- (a) HEMATOCRIT Less than 29%
- (b) HEMOGLOBIN Less than 8 gm/ul
- (c) WBC Less than 4,000 WBC/UL
 Greater than 20,000 WBC/WL
- (d) PT Greater than 15 seconds
 (Coumarin therapy: greater than 30 seconds)
- (e) PTT Greater than 45 seconds
 (Heparin therapy: greater than 70 seconds)
- (f) SODIUM Less than 125 meq/l
 Greater than 155 meq/l
- (g) POTASSIUM Less than 3.0 meq/l
 Greater than 6.0 meq/l
- (h) BUN Greater than 40 mg/dl
- (i) GLUCOSE Less than 50 mg/dl
 Greater than 300 mg/dl
- (j) Positive blood cultures
- (k) Positive CSF cultures
- (l) Positive CSF stains

E. **STEPS:**

1. When a critical value is determined, verify that the specimen was

appropriate and the procedure was correctly performed. Repeat the test if necessary.

2. Notify the module LPO.

3. Notify the ward nursing officer by telephone (or in person).

4. Report.

(a) Patient register number (and full name when available).

(b) Time/date specimen was collected.

(c) Test performed.

(d) Test result including test units (e.g., mg/dl).

5. Record the time, person contacted, and your initials on the back of the lab slip.

F. **RESPONSIBILITY:**

1. Laboratory technician.

(a) Assure correctness of reported value.

(b) Notify appropriate individuals.

2. Ward.

Initiate appropriate action regarding patient treatment.

TAB C-5
FILING LAB SLIPS

A. **PURPOSE:** To provide guidelines for the filing of laboratory slips for pick-up by patient care personnel and for retention in the laboratory.

B. **DEFINITION:** Laboratory slips are multipart standard forms used to request and report laboratory test results.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

Boxes or containers; one for each request location.

D. **CRITERIA:**

1. The original and middle copy of each lab slip will be returned to the requesting location.

2. One box/container will be labeled for each requesting location. These will be placed in the receiving area of the laboratory convenient to patient care staff.

3. The bottom copy of each lab slip will be retained by the laboratory for a period of at least three months, storage space permitting. Lab slips from each section will be filed separately (e.g., hematology separate from chemistry).

4. SF 518's (requests for blood products) will be handled according to "Blood Bank Crossmatch Log/Issuing Blood".

E. **STEPS:**

1. At a minimum boxes are required for:

(a) Wards 1-7.

(b) ICU 1, 2.

(c) Casualty Receiving.

(d) OR Prep & Hold.

(e) Surgical Suite.

(f) Specialty Treatment Area.

2. Separate lab slips into ward and lab copies.

3. File ward copies in the appropriately marked box/container for that request location.

4. File lab copies into empty boxes, alphabetically, by date test was performed.

F. **RESPONSIBILITY:**

1. Laboratory technician:

File lab slips into appropriate boxes/containers for ward pick-up as soon as lab work is complete.

2. Ward personnel.

Periodically check lab boxes for lab slips and deliver to the request center.

3. Module LPO.

Ensure that lab copies are neatly and correctly filed daily, to facilitate easy retrieval.

TAB C-6

CBC - COULTER M-430

A. **PURPOSE:** A suspension of blood cells is passed through a small orifice simultaneously with an electric current. The individual blood cells passing through the orifice introduce an impedance change determined by the size of the cell. The hematocrit is calculated from the RBC pluses by the Analog Card. The leukocyte count and hemoglobin require a lyse reagent to destroy the erythrocytes and convert hemoglobin to a stable pigment while leaving the leukocyte nuclei intact. The hemoglobin is read photometrically. For this facility, a CBC consists simply of a WBC, HGB, and HCT.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Operator's manual.
2. Diluent.
3. Lyse agent.
4. Cleaning agent.
5. Diluent vials.
6. Cell controls.
7. Automatic diluter.

C. **STEPS:**

1. Start up instrument following directions in the operation's manual.
2. Set the dilutor switch to WBC aspirate the sample being tested.
3. Wipe the tip of the aspirator with gauze or a kimwipe. Using a clean vial, dispense the sample, and diluent (WBC dilution).
4. Wipe the tip of the aspirator and change the setting on the diluter to RBC.
5. Aspirate a sample from the WBC dilution just made with the diluter in RBC mode.
6. Wipe the tip of the aspirator and dispense the sample and diluent into a second clean vial (RBC dilution).
7. Place the RBC dilution on the sample platform beneath the RBC aspirator tip and press the start switch.
8. Add 6 drops Lysing solution to the WBC vial. Recap the vial and mix by gentle inversion until the solution is no longer cloudy.
9. Uncap the vial. Place it on the sample platform beneath the WBC aspirator tip and press the start switch.
10. Record the HCT readout (from the RBC dilution) and the WBC and ITGB readouts (from the WBC dilution) that appear.
11. Remove the samples and discard.
12. After completing a run of samples, place vials containing isoton on the

platforms, and sample in the same way as a cell dilution.

13. At the end of a day, cycle cleaning solution through the system.

D. **CONTROLS:**

1. At the beginning of the day, perform manual WBC, HGG, and HCT on a fresh blood sample. Use this as the control for the M430. (If possible, also test a high and a low sample.)

2. Use the same sample as a replicate through out the day.

TAB C-7

WHITE BLOOD CELL COUNT - HEMACYTOMETER

A. **PURPOSE:** A known volume of blood is diluted with 1% glacial acetic acid to a known final volume. The dilute acid lyses the non-nucleated red cells but not the leukocytes or the nucleated red cells. The diluted specimen is delivered into a counting chamber with a known volume and ruled counting area. The cells are allowed to settle and are counted on the basis of the ruled square. The cells counted are multiplied by the calculated dilution factor yielding the white blood cell count.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Hemacytometer.
2. Cover glass.
3. Hand counter.
4. Specimen.
 - a. Whole blood collected in EDTA, heparin, or oxalate.
 - b. Capillary blood.
3. Diluting fluid.
 - a. Glacial acetic acid 1 ml.
 - b. Distilled water QS to 100 ml.

C. **STEPS:**

1. Fill a leukocyte diluting pipette with blood to the 0.5 mark.
2. Carefully remove all excess blood from the outside of the tip of the pipette with a clean gauze. Be careful not to withdraw any blood from the stem on the pipette.
3. While holding the pipette vertically, carefully draw the WBC diluting fluid into the pipette. Gently rotate the pipette during this process to mix the blood and fluid. Be careful not to contaminate the WBC fluid by allowing blood to flow into the fluid.

4. The pipette is full when the fluid reaches the 11 mark.

(a) This results in the following dilution:

0.5 units blood diluted to a final volume of 10 units in the pipette bulb.

$$\frac{0.5}{10.0} = \frac{1}{20}$$

(b) The dilution factor is therefore 20.

5. Repeat steps 1-4 on the same blood sample so that there are two WBC dilutions on that specimen. The specimen must always be run in duplicate.

6. Shake the filled pipettes for about 3 minutes to ensure complete hemolysis of the red cells.

7. Clean the hemacytometer using a lint-free cloth and alcohol or H₂O.
8. The first three drops from the pipette are discarded to eliminate the cell-free fluid from the pipette stem.
9. While holding the pipette vertically, carefully fill one half of the hemacytometer. Do not overfill or underfill. The chamber must fill with one steady movement of fluid.
10. Fill the opposite side of the hemacytometer with the other WBC dilution pipette.
11. Allow the cells to settle, approximately 2-3 minutes, before counting.
12. Scan the hemacytometer to make certain that there is an even distribution of cells.
13. Count the white cells on one half of the hemacytometer using "Low Power" (10X).
14. Count the cells seen in the four large corner squares (1 Sq mm each). Record each result. There should not be more than a 10 cell variation between each of the four squares.
15. Cells on the margin lines.
 - (a) Chambers ruled with double lines: Count the cells that touch the left and upper outer lines. Disregard those cells which touch the right and lower outside lines.
 - (b) Chambers ruled with triple lines: count the cells that touch the middle of the three lines on the left and upper sides. Disregard those cells that touch the middle of the three lines on the lower and right sides.
16. Total the cells counted in the four large corner squares. Calculate the results by multiplying by 50 to give the total WBC per cu mm.
17. Count the cells in the other half of the chamber. Calculate the results and compare with the first half. These must agree within 15% or the test must be repeated.
18. Average the two results. Report the final average.

D. **CALCULATION OF RESULTS:**

1. For each of the white counts performed, calculate the number of WBC/cu mm as follows:
2. Number of WBC X Correction for X Correction for counted (total) Volume Dilution = WBC/cumm
3. Correction for volume will be ten (2) since the chamber is 1/10 mm deep.
4. Correction for dilution will be twenty (2) because the blood was diluted 1:20 as described in step 4. The dilution factor may change if the blood is drawn to a mark other than the 0.5 mark. Those dilution factors must re-calculated as the situation dictates.
5. $10 \times 20 = 200$ However, this number must be divided by four (4) because you counted 4 sq mm.

(a) 200

$$4 = 50$$

6. 50 is a constant multiplication factor when using blood drawn to the 0.5 mark of a WBC pipette.

7. For example: The counts of 4 large corner squares

46
54
52
48
<hr/>
200 total cells

$$200 \times 50 = 10,000 \text{ WBC/cu mm blood}$$

E. **RESULTS:**

Normal values:

<u>Age Group</u>	<u>Range</u>	<u>Average</u>
Normal Infant	8,000-16,500	-
4-7 years	6,000-15,000	10,700
8-18 years	4,500-13,500	8,300
Adult	5,000-10,000	7,000

TAB C-8

MANUAL HEMOGLOBIN DETERMINATION

A. **PURPOSE:** The sahli-hellige method of hemoglobin determination.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Distilled water.

2. Sahli-hellige hemoglobinometer kit containing:

(a) Small bottle of dilute (approx. 0.1N) hydrochloric acid. Prepare this solution by adding 1 ml of concentrated HCl to 99 ml of distilled water. Pour acid into water. Replenish this periodically - it must be of proper strength.

(b) Graduated tube, with a scale on two sides. On one side is the percentage scale, and on the opposite side is the gram scale. The percentage scale reads from 0 to 170. The gram scale reads from 0 to 24.

(c) Pipette, marked at the 20 mm 3 level.

(d) Stirring rod.

(e) Color comparator, with a window in the side. On the right and left sides of this opening is the color standard for comparison. The center has an open slot to hold the graduated tube.

C. **STEPS:**

1. With a medicine dropper, place five drops of the 0.1 N HCL in the bottom of the graduated tube. Place tube in color comparator.

2. Using well-mixed venous blood or fingertip blood, fill pipette to the 20 mm 3 mark.

3. Wipe blood from the outside of the pipette. Transfer blood to Sahli tube. Note time.

4. Aspirate distilled water into pipette two or three times and transfer these washings to tube.

5. Shake until blood is well mixed and the tube is a uniform color.

6. Add distilled water, drop by drop, each time mixing the solution with the stirring rod. Keep adding water and mixing until the color of the solution matches the standards on either side. Remove stirring rod from the tube each time before comparing. Natural light makes more accurate readings possible.

7. Five minutes after time is noted, read the result from the scale on the tube by noting the graduation mark at the lower edge of the meniscus.

D. **RESULTS:**

1. Report the grams of hgb per 100 ml of whole blood.

2. In the Navy kit, 100% is equal to 14.5 gm/ml. If either scale is hard to read, compute the other value. (100% = 14.5gm; 6.9% = 1 gm.)

TAB C-9

MANUAL HEMATOCRIT

A. **PURPOSE:** Hematocrit is the volume of red blood cells expressed as a percentage of the value of whole blood in a sample. Whole blood is anticoagulated, and the total value of the red cell mass is expressed as a percentage of the whole blood volume.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Capillary tubes, 70x1 mm (heparinized if capillary blood is used).
2. Microhematocrit centrifuge.
3. Hematocrit reader.
4. Tube sealer (modeling clay).

C. **STEPS:**

1. Fill two capillary tubes to within 10-15 mm of the end with capillary or well-mixed whole blood (EDTA anticoagulated).

2. Seal the dry end of the tube by inserting it into the tube sealer.

3. Place the sealed capillary tubes containing the blood sample in the head of the centrifuge with the sealed ends toward the periphery and against the elastic strip. It is important that an even number of tubes be used and one-half placed in opposition to the other half for proper balance.

4. Close the cover and push the latch until it is locked. Turn the timer knob clockwise, past the 5, until a click is heard; then turn the knob back to 5.

5. After the centrifuge stops, open the centrifuge, and remove the head cover and remove the capillary tubes.

6. Read the percentage of red blood cells from the microhematocrit reader.

(a) Place the capillary tube in the groove of the plastic indicator so that the bottom of the packed red cells column coincides with the black line on the bottom of the plastic indicator.

(b) Rotate the scale plate until the top of the plasma column coincides with the 100% beneath the red line on the plastic indicator.

(c) Rotate both discs together until the spiral line intersects the capillary tube at the red and white cell interface.

(d) Read the hematocrit from the point on the scale directly beneath the red line of the indicator.

D. **RESULTS:**

1. Normal values.

	RANGE (%)	Average (%)
Male	42-52	47
Female	37-47	42

Newborn	44-64	.54
1-y.o		35
10-y.o		37.5

2. Controls.

(a) Specimens must be run in duplicate and must agree within ± 0.01 .

(b) A patient replicate sample can be run periodically throughout the day as an added control.

(c) To compare against the coulter, multiply the spun HCT value $\times 0.98\%$. This should agree within 3% of the coulter value.

(d) The maximum packing time should be determined before using the microhematocrit centrifuge for patient samples.

E. MAXIMUM PACKING TIME:

1. Fill 12 microhematocrit capillary tubes.
2. Spin first pair for 1 minute.
3. Spin second pair for 2 minutes.
4. Spin third pair for 3 minutes.
5. Spin fourth pair for 4 minutes.
6. Spin fifth pair for 5 minutes.
7. Spin sixth pair for 6 minutes.
8. Record pack cell volume for the microhematocrit reader for each pair.
9. Select centrifuging time where values are the same. Take longer time as the setting:

Example:

1 min	=	37
2 min	=	36
3 min	=	35
4 min	=	35 - Choose this time
5 min	=	35

TAB C-10

PERIPHERAL SMEAR PREPARATION

A. **PURPOSE:** It is important to make a satisfactory smear for evaluation since much information is gathered from it's examination.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Glass slides.
2. Capillary, or blood drawn in EDTA.
3. Wooden applicator sticks.

C. **STEPS:**

1. Place a small drop of blood about 3/4" from the end of a clean glass slide, and lay the slide on a flat surface.
2. Grasp another slide between thumb and index finger of your right hand. Place the end of the spreader slide on the lower slide, just ahead of the drop of blood and at an angle of about 30-40.
3. Pull the spreader slide back to the edge of the blood and allow the blood to spread out between the two surfaces.
4. Then the blood has spread along two-thirds of the width, push the spreader slide forward in a steady, even motion. (The film should not touch the margins of the slide.)
5. Allow the smear to air dry.
6. Label the smear with the patients' name or number.

D. **RESULTS:**

1. A satisfactory blood film should have:
 - (a) A thick portion and a thin portion with a gradual transition from one to the other.
 - (b) A smooth, even appearance.
 - (c) No ridges, waves or holes.
2. The thickness of the film can be regulated by:
 - (a) Changing the angle at which the spreader is held.
 - (b) Varying the pressure and speed of spreading.
 - (c) Using a smaller or larger drop of blood.

TAB C-11

WRIGHT'S STAIN

A. **PURPOSE:** Wright's stain is a mixture of eosin and methylene blue used for the preparation of blood smears for microscopic examination.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Methanol.
2. Wright stain.

C. **STEPS:**

1. Fix smear in methanol.
2. Place dry, fixed smear on staining rack.
3. Flood smear with about 1 ml of wright's stain and allow it to stand for 2 minutes.
4. Add an equal amount of buffer.
5. Gently blow on the surface of the slide to mix the buffer and the stain. Mix until a metallic film appears.
6. Let stand for 3 minutes.
7. Wash with tap water (with neutral PH, otherwise use distilled water). Be sure to float the metallic film off the slide to prevent streaking.
8. Remove stain from the bottom of the slide. Let air-dry.

D. **RESULTS:**

1. Check slide under a microscope for proper color.
2. Vary staining times accordingly.

TAB C-12

LEUKOCYTE DIFFERENTIAL

A. **PURPOSE:** The leukocyte differential consists of a systematic evaluation and classification of leukocytes on a wright's stained peripheral blood smear, expressing in percentage the relative number of the various types of leukocytes present. In addition, the morphology of the erythrocytes and platelets and an estimate of platelet adequacy are noted.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Microscope with oil immersion lens.
2. Immersion oil.
3. Diff counter.
4. Lens paper.

C. **STEPS:** Leukocyte classification.

1. Inspect smear under 45X power for cell distribution and staining. Remake or re-stain unsatisfactory smears as necessary.

2. Select the area to be examined near the feathered edge of the smear where the edges of the erythrocytes are nearly touching one another.

3. Estimate the total leukocyte count by examining several high-dry fields.

<u>Average No. Leukocytes/h.p.f.</u>	<u>Estimated Total Leukocyte Count (in thousands)</u>
2-4	4-7
4-6	7-10
6-10	10-13
10-20	13-18

4. Perform leukocyte differential under oil immersion.

(a) Move slide from the upper edge of the smear to the extreme lower edge, counting and classifying each leukocyte in the successive fields.

(b) Shift over one field and proceed to the upper edge, continuing to classify each cell.

(c) Continue in this fashion until 100 cells are counted.

(d) Count only intact leukocytes. Classify all other cells encountered, obtaining consultation from experienced personnel when needed.

(e) For leukocyte counts less than 1,500/mm, perform a 50-cell differential. Make a note on the report that 50 cells were enumerated. Do not multiply by 2 as this will make it appear that 100 cells are classified. Report percentage of each type of leukocyte.

5. Note presence of any morphologic abnormalities of the leukocytes, such as:

(a) Hyper-segmentation of the nucleus of neutrophils (report percentage).

(b) Toxic granulation of neutrophils, nuclear degeneration, vacuolization and degeneration of the cytoplasm, and Dohle bodies (report, e.g., 1+-4+).

(c) Atypical lymphocytes (report percentage).

D. **STEPS:** Platelet estimate.

1. Under oil immersion, examine several fields where the RBC's are touching (but not overlapping).

2. Count the average number of platelets per field. Calculation:
Estimated No. Platelets/mm = No. Platelets/O.I.F.x 20,000.

3. Report platelets as adequate, increased, or decreased. A rough estimate may be made as follows:

Less than 6/h.p.f.	decreased
Several with occasional clumps	adequate
More than 20/h.p.f.	increased

4. Note abnormal variation in size and morphology of platelets (e.g., "giant" or atypical platelets).

5. In cases of abnormally high or low WBC counts do the following:

(a) Count the number of platelets per 1000 RBC's.

(b) Multiply by the RBC count.

(c) Divide by 1000.

(d) This equals the platelet count per CU mm and is more accurate in these cases.

E. **STEPS:** Erythrocyte morphology.

1. Under oil immersion, carefully examine erythrocytes in an area where they are touching (but not overlapping).

2. Grade findings on a 1+-4+ scale as follows:

20%	slight or rare/h.p.f.	1+
50%	moderate number/h.p.f.	2+
75%	many/h.p.f.	3+
90-95%	uniform involvement	4+

3. Terms used.

Anisocytosis

Variation in size; classify microcytosis and macrocytosis and degree of both (use eyepiece micrometer for accuracy).

Poikilocytosis

Variation in shape; classify (e.g. bizarre, elliptocytosis, target cells,

sickle cells, burr, teardrop).

Hemoglobin content

Note hypochromia; (hyperchromia is Erythrocytic inclusions nonexistent except in hereditary spherocytosis). Stippling, howell-jolly bodies, parasites, Cabot rings, nonspecific basophilic inclusions.

Miscellaneous

Polychromatophilia; Rouleaux formation; nucleated red blood cells (NRBC's).

4. Report NRBC's as the number counted per 100 leukocytes. If more than 5 NRBC's per 100 WBC's are counted, correct the total leukocyte count.

$$\text{Corrected WBC Count} = \frac{\text{Uncorrected WBC}}{\text{NRBC's} + 100} \times 100$$

F. **RESULTS:**

1. The normal percentage distribution of WBC's is:

Bands	0-5
Segs	56-62
EOS	0-3
Basos	0-1
Lymphs	20-30
Monos	0-8

G. **CARE OF SLIDES:**

1. Gently wipe oil from the slide using lens tissue.
2. File for future reference.

TAB C-13

CEREBROSPINAL FLUID (CSF) CELL COUNT

A. **PURPOSE:** CSF is examined in a counting chamber and the number and type of cells are noted.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. CSF from the third collection tube (least likely contaminated with peripheral blood).

2. WBC diluent.

(a) Gentian violet, 1% 1 ml

(b) Glacial acetic acid 1 ml

(c) Distilled water: q.s. to 100 ml

3. RBC diluent: isotonic saline.

4. Hydrochloric acid, 0.1 N.

5. RBC and WBC diluting pipette.

6. Hemocytometer with coverslip.

7. Microscope.

8. Mixer.

C. **STEPS:** Describe color and gross appearance of fluid.

1. Clear: Perform the cell count without the use of diluting fluid. Count and differentiate the cells within the nine large squares on each side of the hemocytometer.

Calculation: No. cells/mm³ = Cells counted x $\frac{10}{18}$

18

2. Slightly turbid: WBC count.

(a) Fill a clean WBC diluting pipette to the 0.5 mark with WBC diluent.

(b) Aspirate the well-mixed CSF to the 11 mark.

(c) Mix on a mechanical mixer for 5 minutes.

(d) Discard the first three drops from the pipette.

(e) Charge both chambers of the hemocytometer.

(f) Allow the cells to settle for one minute.

(g) Count and differentiate PMN's, mononuclear WBC's, and unidentified cell types within the nine large squares on each side of the hemocytometer.

Calculation: No. cells/mm³ = No. cells counted (both sides) x 0.58.

3. Visibly bloody.

(a) WBC count.

- (1) Aspirate the well-mixed CSF to the 1.0 mark of a WBC pipette.
- (2) Aspirate 0.1 N HCL to the 11 mark.
- (3) Mix on a mechanical mixer for 5 minutes.
- (4) Load the hemocytometer.
- (5) Allow cells to settle for one minute within four large squares one each side of the hemocytometer.
- (6) Count and differentiate PMN's, mononuclear WBC's, and unidentifiable cells.

Calculation:

No. cells/mm³ - No. Cells (both sides) x 12.5.

(b) RBC count.

- (1) Aspirate the well-mixed CSF to the 1.0 mark of a RBC pipette.
- (2) Aspirate RBC diluent to the 101 mark.
- (3) Mix on a mechanical mixer for 5 minutes.
- (4) Fill both counting chambers of the hemocytometer.
- (5) Allow cells to settle for one minute.
- (6) Count the number of RBC's within the five small squares of the center large square on both sides of the hemocytometer.

Calculation:

NO. RBC's/mm³ = No. cells counted (both sides x 2,500.
Sic: 50 RBC's counted x 2,500 = 125,000 RBC's/mm³.

D. **RESULTS:**

Normal values.

<u>RBC's</u>	<u>LYMPHOCYTES</u>	
Adults	None	0-08
Newborns	None	0-30

TAB C-14

BODY FLUID CELL COUNT

A. **PURPOSE:** Various body fluids are examined on a hemocytometer and the number of cells are noted. Leukocyte differentiation is performed on a smear, stained with wright's stain. The fluid cell count and differential are useful in diagnosing a number of conditions when used in conjunction with the results of various chemical and bacteriological tests.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Body fluid.
2. Fluid diluent: 0.1% methylene blue in normal saline (0.1 gm methylene blue in 100 ml saline).
3. Red cell diluent:
 - (a) Combine 12.5g sodium sulfate with 33.3 ml glacial acetic acid.
 - (b) Distilled water: Qs to 200 ml.
4. Hydrochloric acid 1/10 N.
 - (a) Add 8.6 ml concentrated hcl to distilled water.
 - (b) Qs to 1,000 ml.
5. RBC and WBC diluting pipettes.
6. Hemocytometer with coverslip.
7. Microscope.
8. Mixer.

C. **STEPS:**

1. Describe the color and general appearance of the fluid.
2. Thoroughly mix the sample and draw up to the 1.0 mark of the WBC pipette.
3. Draw fluid diluent up to the 11 mark.
4. Thoroughly mix on a mechanical mixer for 3 minutes.
5. Count and differentiate the cells on the hemocytometer.

Calculation: $\text{No. Cells/cu mm} = \text{cells counted} \times 10 \times \frac{10}{18}$

6. When numerous RBC's are present, proceed as follows:
 - (a) WBC count.
 - (1) Draw sample to the 1.0 mark of a WBC pipette and 1/10 N HCl to the 11 mark.
 - (2) Count the WBC's in the four large squares.

Calculation: $\text{WBC's/cu mm} = \text{No. WBC's counted} \times 25.$

(b) RBC count.

(1) Draw sample to the 1.0 mark of a RBC pipette and RBC diluent to the 101 mark.

(2) Count the RBC's in the five small squares in the center RBC area.

Calculation: $\text{RBC's/cu mm} = \text{No. RBC's Counted} \times 5,000.$

D. **RESULTS:**

Normal values.

	<u>SYNOVIAL FLUID</u>	<u>PLEURAL FLUID</u>
Leukocytes	Less than 200 cells/cu mm	Less than 100 cells/cu mm
Differential	Less than 25% graunlocytes	Less than 25% graunlocytes

TAB C-15

MALARIAL SMEAR

A. **PURPOSE:** To provide guidelines for the preparation of thick and thin blood smears to look for malarial parasites.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Blood from a purple top tube (EDTA).
2. Wright - giemsa stain.
3. Glass slides.

C. **STEPS:**

1. Place a large drop of blood on a glass slide and spread, using the corner of another slide, to cover an area about the size of a dime.
2. Air dry for 30 minutes.
3. Prepare a thin smear (as for a differential).
4. Stain both smears with Wright-giemsa stain following the manufacturer's instructions.
5. Examine under oil immersion.

D. **RESULTS:**

1. Carefully examine both thick and thin smears for material parasites. Parasites stain varying shades of blue, pink, and red.
2. Report as "No material parasites seen" if negative findings.
3. If positive, confirm with lab officer before reporting.

TAB C-16

AUTOMATED PT/PTT

A. **PURPOSE:** To provide guidelines for performing PT (prothrombin time) and PTT (partial thromboplastin time) tests on the Electra 750 coagulation analyzer.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Electra 750.
2. 0.1 ml pipette with tips.
3. Test cuvettes.
4. 2 reagent cups fitted with 0.1 ml tubing.
5. Thromboplastin reagent.
6. APTT reagent.
7. Calcium chloride solution.
8. Control plasmas.

C. **CRITERIA:**

1. Tests will be run in duplicate.
2. Controls will be run every day.

D. **STEPS:**

1. PT (Prothrombin Time).

(a) Set up instrument as detailed in operator's manual.

(b) Reconstitute reagents and controls according to manufacturer's instructions. Place reagents in reagent cups. Use magnetic stirrer for thromboplastin reagents.

(c) Press PT 1 or PT 2 (for single-or dual-reagent testing, respectively).

(d) Press START twice within three seconds to initiate confidence test. Compare times to previous confidence test time.

(e) Pipette 0.1 ml of control plasma into each well of PT cuvette. Pipette 0.1 ml patient plasma to be tested into cuvette (one side for single test, both sides for dual tests).

(f) Load cuvettes into refrigerated turntable positions, observing cuvette directionality.

(g) Prime pumps.

(h) Set AUTOMATIC/MANUAL switch to AUTOMATIC position.

(i) Press START. Instrument will proceed with all tests under automatic control.

(j) Continue to load sample cuvettes into turntable as slots become available.

(k) Record data.

2. Empty cuvette catch bin when test run is finished.

3. PTT (Partial Thromboplastin Time).

(a) Set up instrument as in operator's manual.

(b) Reconstitute reagents and controls according to manufacturer's instructions. Place reagents in reagent cups. Use magnetic stirrers for APTT reagents.

(c) Press APTT.

(d) Press START twice within three seconds to initiate confidence test. Compare times to previous confidence test times.

(e) Pipette 0.1 ml of control plasma or patient plasma into each well of an APTT cuvette, either singly or two samples per cuvette.

(f) Load cuvettes into refrigerated turntable positions, observing cuvette directionality.

(g) Prime pumps.

(h) Set AUTOMATIC/MANUAL switch to AUTOMATIC position.

(i) Press START.

(j) Continue to load sample cuvettes into turntable as slots become available.

(k) Record data.

(l) Empty cuvette catch bin when test run is finished.

TAB C-17

PROTHROMBIN TIME (PT) - TILT TUBE METHOD

A. **PURPOSE:** To provide guidelines for the performance of a prothrombin time using the tilt tube method. This method will be utilized as the backup, should the automated coagulation analyzers fail.

B. **EQUIPMENT, SUPPLIES, AND FORMS NEEDED:**

1. Freshly drawn blue top tube.
 - (a) Centrifuge sample.
 - (b) Separate plasma from cells immediately.
 - (c) Keep plasma refrigerated.
2. Reconstitute and store reagents according to package insert.
 - (a) Thromboplastin reagent.
 - (b) Calcium chloride.
 - (c) Coagulation Controls.

C. **STEPS:**

1. Mix one part thromboplastin with one part calcium chloride (0.025m) and place into a 37°C water bath to warm.
2. Pipette 0.1 ml test plasma into a 12 x 75 mm tube immediately prior to testing.
3. Place tube into a 37°C water bath for 1 minute.
4. At 1 minute, pipette 0.2 ml of the thromboplastin - calcium chloride mixture into the test plasma and start a stopwatch.
5. The clotting time is determined by observing the appearance of the first fibrin strands in the reaction mixture while tilting the tube so that the mixture runs about halfway up the side.
6. Repeat in triplicate and average the results.

D. **RESULTS:**

1. A normal plasma will give a reading between 12 and 16 seconds.
2. Note that times will be slightly longer by this method than by other methods due to technician participation.

TAB C-18

ACTIVATED PARTIAL THROMBOPLASTIN TIME (APTT) - TILT TUBE METHOD

A. **PURPOSE:** To provide guidelines for the performance of an activated partial thromboplastin time using the tile tube method. This method will be utilized as the backup, should the automated coagulation analyzer fail.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Freshly drawn blue top tube.
 - (a) Centrifuge sample.
 - (b) Separate plasma from cells immediately.
 - (c) Keep plasma refrigerated.
2. Reconstitute and store reagents according to package inserts.
 - (a) APTT reagent.
 - (b) Calcium chloride.
 - (c) Coagulation controls.

C. **STEPS:**

1. Mix one part APTT reagent with one part calcium chloride (0.025m) and place into a 37°C water bath to warm.
2. Pipette 0.1 test plasma into a 12 x 75 mm tube immediately prior to testing.
3. Place tube into a 37°C water bath for 1 minute.
4. At 1 minute, pipette 0.2 ml of the APPT - calcium chloride mixture into the test plasma and start a stopwatch.
5. The clotting time is determined by observing the appearance of the first fibrin strands in the reaction mixture while tilting the tube so that the mixture runs about half way up the side.
6. Repeat in triplicate and average the results.

D. **RESULTS:**

1. A normal plasma will give a reading between 30 and 40 seconds.
2. Note that times will be slightly longer by this method than by other methods due to technician participation.

TAB C-19

FIBRINOGEN DETERMINATION

A. **PURPOSE:** The test is based on the quantitative fibrinogen assay, developed by Clauss, in which the clotting time of dilute plasma is measured upon the addition of thrombin. At high thrombin concentrations, the reaction rate is determined by the fibrinogen concentration. The clotting time observed is then compared with that of a standardized fibrinogen preparation.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Freshly drawn blood (9 parts) to 3.8% sodium citrate (1 part); blood drawn in a blue top tube.

(a) Centrifuge sample.

(b) Separate plasma from cells immediately.

(c) Keep plasma refrigerated.

2. Reconstitute and store reagents according to package insert.

(a) Thrombin reagent.

(b) Fibrinogen calibration reference: Prepare calibration curve according to package insert.

(c) Owren's veronal buffer.

(d) Normal plasma control.

C. **STEPS:**

1. Add 0.1 ml patient plasma to 0.9 ml owren's veronal buffer.

2. Add 0.1 ml control plasma to 0.9 ml owren's veronal buffer.

3. Perform duplicate determinations on each dilution by following the electra 750 operator's manual.

D. **RESULTS:**

1. Very high fibrinogen values of greater than 800 mg/dl:

(a) Dilute plasma 1:20 (0.1 ml plasma to 1.9 ml owren's veronal buffer equals 1:20 dilution).

(b) Obtain value from calibration curve and multiple by 2.

2. Very low fibrinogen values of less than 40 mg/dl:

(a) Dilute the plasma 1:5 (0.2 ml plasma to 0.8 ml owren's veronal buffer).

(b) Repeat the test.

(c) Obtain value from the calibration curve and divide by 2.

3. Normal value: 150-400 mg/dl.

TAB C-20

URINALYSIS

A. PURPOSE:

1. The physical and chemical properties of normal urine are markedly constant; any abnormalities are easily detected. The use of simple tests provides the physician with helpful information concerning the diagnosis and management of many diseases.

2. A routine urinalysis includes color, clarity, chemistries, specific gravity, and microscopic examination.

B. EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:

1. Freshly voided urine specimens. If testing cannot be performed within an hour after collection, the specimen must be refrigerated to 2° to 8°C and returned to room temperature before testing.

2. N-multistix.

3. Urine test tube.

4. Refractometer.

5. Centrifuge.

6. Microscope.

C. STEPS:

1. Record the color and clarity of the specimen.

2. Ensure stage is dry.

3. Place one drop of urine under the cover plate of the refractometer.

4. Hold instrument up to light source.

5. Read scale at light-dark boundary.

6. Record the specific gravity.

7. Wipe stage dry with a kimwipe.

8. Briefly (no longer than 1 second) dip test strip into the urine. Make sure all reagents are totally immersed.

9. Remove excess urine by drawing the edge of the strip along the rim of the container.

10. After an appropriate time, as indicated below, read the test as follows:

HOLD STRIP CLOSE TO COLOR BLOCKS AND MATCH CAREFULLY.

PH	Immediate
Protein	30-60 seconds
Glucose	30-60 seconds
Ketones	60 seconds

Bilirubin	30-60 seconds
Blood	60 seconds
Urobilinogen	10-30 seconds
Nitrite	30 seconds

For convenience, all values on the strip may be read at 60 seconds (during the second minute) after immersion in the urine. Color changes that occur after 2 minutes from immersion are not of diagnostic value. Color changes that occur only along the edge of the test area should be ignored. Careful removal of excess urine should eliminate this effect.

11. Pour 10-15 ml of mixed specimen into a urine test tube.

(NOTE: microscopics are performed only if a chemistry is positive, or upon specific result.)

12. Centrifuge at 2000 RPM for 5 minutes.

13. Pour off supernatant fluid.

14. Re-suspend the sediment in the urine that drains back down the sides of the tube.

15. Place one drop on a microscope slide.

16. Cover with a coverslip and examine on high dry immediately.

D. RESULTS:

<u>1. Color</u>	<u>Possible Cause</u>
Straw to amber	Normal
Orange	Concentrated urine
Deep yellow	Riboflavin
Bright orange	Pyridium
Orange brown	Urobilin
Greenish orange	Bilirubin
Smokey	RBC's
Wine red or reddish brown	HBG pigments
Brown to black on standing	Melanin or hemo
Almost colorless	Diluted urine
Reddish orange, alkaline	Rhubarb
Dirty green on standing	Excess indican
Red in alkaline solution	Phenolythalein
Green or blue	Methyline blue
Greenish yellow florescence	Some vitamins

2. Reaction. Acid or alkaline - 6.0 - normal mark.

3. Specific gravity. 1.010 - 1.025 - considered normal.

4. Proteins. Detectable amounts of protein are not normally present.

5. Glucose. Negative result is normal, detectable amount is indicative of something.

6. Bilirubin. Negative result is normal, detectable amount is indicative of something.

7. Nitrite. Positive result is indicative of bladder infection.

8. Follow the uniform standard of reporting microscopic findings:

(a) Scan, under low power objective, at least 10 fields. Count and average the counts.

(b) Scan, under high power objective, at least 10 fields. Count and report the average range of the following elements:

<u>ELEMENTS</u>	<u>REPORT</u>
Casts:	Range/LPF
White Blood Cells:	Range/HPF (If more than 100, report more than 100/HPF)
Red Blood Cells:	Range/HPF
Crystals:	Few, moderate, many & type of crystal
Amorphous:	Few, moderate, many
Epithelial Cells:	Few, moderate, many
Bacteria:	Few, Moderate many
Trichomonas:	Present
Yeast:	Few, moderate, many; report mycelial phase if present
Mucus:	Few, moderate many
Miscellaneous:	Use your judgement

9. Normals.

(a) Casts, hyaline: 0-1/LPF; all other casts should not be seen.

(b) White Blood Cells: 0-5/HPF.

(c) Red Blood Cells: 0-3/HPF.

(d) Crystals: Presence usually not significant unless found persistently in patients with renal calculi or if associated with certain metabolic diseases; i.e., sulfonamides, cystine, leucine and tyrosine crystals.

(e) Amorphous: Present in urines that have been left standing for awhile.

(f) Epithelial cells: 0-4/HPF.

(g) Bacteria: May be present unless specimen is catheterized or freshly voided in a clean container.

(h) Mucus: Presence not significant.

(i) Large numbers of erythrocytes, leukocytes and casts may appear in the urine of healthy subjects who perform strenuous exercise or who are exposed to serve cold. Except under these conditions, certain abnormal constituents always indicate renal disease (e.g., red cell casts and white cell casts).

TAB C-21

PREGNANCY TEST

- A. **PURPOSE:** To provide guidelines for the performance of the pregnancy test.
- B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**
 - 1. Pregnancy test kit.
 - 2. Urine (preferably first morning void).
- C. **STEPS:**
 - 1. Perform test according to package insert.
 - 2. Run positive control along with patient specimen.
- D. **RESULTS:**

Report as either positive or negative.

TAB C-22

MONONUCLEOSIS TEST

A. **PURPOSE:** To provide instructions for performance of the screening test for mononucleosis.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

Monospot slide test kit (FSN 6506-01-005-4375).

D. **CRITERIA:**

1. Serum or plasma.
2. A positive and negative control must be run along with patient samples.

E. **STEPS:**

1. On a clean slide (supplied with kit) place one drop of guinea pig antigen, reagent I, into box number 1.

2. Place one drop of the beef erythrocyte stromata, reagent II, into box number 2.

3. Add one drop of test serum or plasma to both boxes. Mix each with separate sticks.

4. Add one drop of horse erythrocyte antigen (supplied with kit) to both boxes. Mix each with separate disposable sticks.

5. Rock slide back and forth for 2 minutes so that liquid flows slowly over the entire area of the boxes.

6. Read results in 2 minutes.

(a) Agglutination in box 1 is positive for infectious mononucleosis.

(b) No agglutination in either box is negative for mononucleosis.

TAB C-23

RPR (RAPID PLASMA REAGIN CARD TEST FOR SYPHILIS)

- A. **PURPOSE:** To provide instructions for the performance of an RPR test.
- B. **DEFINITION:** The RPR test is a sensitive, easily performed, screening test for syphilis.
- C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**
1. RPR Kit (FSN 6550-00-159-5011).
 2. Rotator.
- D. **CRITERIA:**
1. Performed on unheated plasma or serum.
 2. A positive and negative control must be run along with patient samples.
- E. **PREPARATION OF ANTIGEN:**
1. Attach needle hub to tapered fitting on plastic dispensing bottle.
 2. Gently shake antigen ampule to suspend particles and then snap off top at the break line on the neck of the ampule.
 3. Withdraw the antigen suspension into the collapsible dispensing bottle by suction. The antigen is good for 1 month without and 3 months with refrigeration.
 4. Upon completing the test, remove the needle, clean it, and replace the screw cap on the dispensing bottle.
- F. **PERFORMING THE TEST:**
1. Draw the plasma or serum up into the dispenstir.
 2. Place one drop of the plasma or serum into one of the test circles of the diagnostic card.
 3. Shake the antigen dispensing bottle gently, holding it in a vertical position while squeezing a drop of the antigen onto the test area.
 4. Use the flat end of the dispenstir mix the antigen suspension with the test suspension and spread the mixture over the entire area.
 5. Rotate the card at the speed specified in the product insert for a full 8 minutes.
- G. **READING AND REPORTING THE RESULTS:**
1. Read immediately under a bright light for flocculation or agglutination.
 2. Report test as:
 - (a) Reactive if specimen shows agglutination or flocculation.
 - (b) Nonreactive if specimen shows no agglutination.

TAB C-24

PLANTING CULTURES

A. **PURPOSE:** Cultures are planted according to source onto nutrient agar and selective media in a fashion as to isolate probable pathogens for further identification and susceptibility testing.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. 1:1000 inoculating loop.
2. BAP (Blood agar plate).
3. MAC (MacConkey agar).
4. Choc (Chocolate agar).
5. T.M. (Thayer-Martin agar).
6. Blood culture bottles.
7. Thioglycollate broth (thio).
8. Inoculating loop.
9. SS agar.
10. GN broth.

C. **STEPS:**

1. Streak inoculum for isolation according to the following guidelines.
 - (a) Urine - use 1:1000 loop; BAP, MAC, smear for gram stain.
 - (b) G.C. Screen - T.M. plate (Thayer-Martin agar), Choc, smear for gram stain.
 - (c) Throats for strep screen - BAP.
 - (d) Wound - BAP, Choc, Thio; MAC; smear - for gram stain.
 - (e) Stool - MAC, SS Agar, GN broth.
 - (f) CSF - BAP, Choc, MAC, Thio, smear for gram stain.
 - (g) Blood - blood culture bottles.
 - (h) Others - treat like a wound or consult your supervisor.
2. Incubate all MAC and THIO in the room air side of the 37⁰C incubator. Incubate all other media in the CO₂ side of the incubator.

D. **RESULTS:**

Work up plates according to specimen source.

TAB C-25

GRAM STAIN

A. **PURPOSE:** The mechanism of the gram stain is not clearly understood. There is general agreement that a gram-positive organism retains the primary stain after decolorization due to a variety of factor including an isoelectric point at ph2, presence of a magnesium ribonuclease protein complex, a phosphoric ester and the mordant effect of iodine. Organisms are judged to be gram positive if they retain the primary stain after decolorization. Gram-negative organisms are decolorized and appear pink to red because they take up the counterstain.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Aqueous crystal violet.
2. Decolorizer, acetone-isopropyl alcohol (25/75 v/v).
3. Gram's iodine.
4. Safranin, alcoholic.

C. **STEPS:**

1. Place cooled heat-fixed specimen slide into crystal violet stain for one minute.
2. Remove slide and rinse with cool water.
3. Remove excess water and place slide into Gram's iodine for one (1) minute.
4. Remove slide and rise with cool water.
5. Flood off excess water with decolorizer until solvent runs colorlessly from slide. Caution: Do not over de-colorize.
6. Wash slide with tap water and place into safranin for one minute.
7. Remove slide, wash with cool tap water, blot dry using blotting paper, and examine using oil immersion lens.

D. **RESULTS:**

Retention of the purple-black mordant treated primary stain indicates a gram-positive microorganism. Microbial cells which decolorize and stain pink to red with the counterstain are gram negative.

TAB C-26

CATALASE TEST

A. **PURPOSE:** Staphylococci produce the enzyme catalase which when mixed with hydrogen peroxide will liberate oxygen from hydrogen peroxide with the occurrence of vigorous bubbling.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

Hydrogen peroxide (H_2O_2) 3.0%.

C. **STEPS:**

1. Place one drop of H_2O_2 on a clean glass slide.
2. Pick the colony in question from an agar plate by means of a cooled (after flaming red hot) wire loop. Sterile applicator sticks are satisfactory.
3. Dip the loop containing the colonies of bacteria in the drop of H_2O_2 and observe for immediate vigorous bubbling.

D. **RESULTS:**

1. Immediate bubbling denotes a positive test.
2. Staph spp. and Niesseria Spps. are catalase positive. Strep & nheumococci are catalase - negative.
3. Avoid digging the wire loop or applicator stick into the BAP.
4. Platinum wire may produce false positive reaction.

TAB C-27

COAGULASE TEST

A. **PURPOSE:** Coagulase activity is essentially confined to staphylococci and is related to the pathogenic species. It is demonstrated when a SSP of staph capable of producing the enzyme coagulase is added to human or rabbit plasma and a clot is formed. This procedure can be performed on a glass slide or in a glass test tube.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Plasma-human or rabbit-fresh or rehydrated.
2. Normal saline.

C. **STEPS** - Slide method.

1. Place a drop of normal saline on a clean glass slide and prepare a rather heavy, even suspension of staph in it.
2. Place a loopful of fresh or recently reconstituted dehydrated plasma in the suspension. Mix and then withdraw the loop.
3. Immediately observe for the formation of a clot. This usually occurs within a few seconds with coagulase positive SSP.
4. A negative slide test must be confirmed by a tube test.

D. **STEPS** - Test tube method.

1. Transfer 0.5 ml of a 24 hour broth culture of staph or transfer a large loopful of growth from an agar plate to 0.5 ml of human or rabbit plasma in a glass tube.
2. Incubate the tube at 37°C preferably in the heatblock for 3 hours. Observe approx. every 30 minutes for clotting. If negative after 3 hours, re-check reading after 24 hours incubation.

E. **RESULTS:**

1. Pathogenic spp. of staph usually gives a positive reaction (clot formation).
2. Negative reaction-suspension remains homogenous and clot formation does not take place.
3. When colonies of gram positive calli with zones of complete hemolysis suspected of being Saurveus are tested, most catalase +, coagulase + colonies are Saureus.

TAB C-28

OXIDASE TEST

A. **PURPOSE:** This test is based on the production of an oxidase enzyme by the members of the genera *Neisseria* and *Pseudomonas*. Upon the addition of oxidase reagent to growth of an organism in question, the presence of oxidase enzyme is evidenced by a color change in the colonies.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Taxo N-discs - dimethyl-p-phenylenediamine Hydrochloride.
2. Tetramethyl-p-phenylenediamine HCL.

C. **STEPS:**

1. Place an n-disc on a clean glass slide and moisten with a drop of distilled water.
2. Apply suspicious colonies or growth from media to the moistened disc using sterile technique.

D. **RESULTS:**

1. Colonies of oxidase-positive organisms first become pink, then maroon, through purple to black.
2. A positive oxidase test together with the finding of gram negative diplococci in gram-stained smears will differentiate *Neisseria* from other organisms; if cultured on T-M agar or transgrow medium, the reaction constitutes a presumptive positive test for *Neisseria gonorrhea*.
3. Oxidase test is intended only as an aid in the identification of *Neisseria* and *Pseudomonas*.

TAB C-29

FECAL SCREEN FOR OCCULT BLOOD

A. **PURPOSE:** Hemoglobin exerts a peroxidase-like activity and facilitates the oxidation of alpha guaiaconic acid, a phenolic compound, by hydrogen peroxide. This oxidation reaction turns the guaiac paper blue within 30 seconds. Positive results are expected in ulcer, hiatal hernia, or colon rectal cancer patients.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Small stool sample.
2. Occult blood kit with developer.
3. Water.

C. **STEPS:**

1. Open slide and apply thin stool smear inside boxes.
2. Open perforated window in back of slide.
3. Apply 2 drops of developer to paper directly over each smear.
4. Record results after 30 seconds. Any trace of blue is positive for occult blood.
5. Results are verified by applying one drop of developer between the positive and negative performance monitors. Make sure they test positive and negative as marked. If not, repeat the test on another card.

D. **RESULTS:**

Report negative or positive for occult blood.

TAB C-30

SPORE STRIPS

A. **PURPOSE:** Spore strips are used to determine if steam autoclaves are functioning properly. Spore strips are autoclaved as usual and delivered to the laboratory for growing.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Nutrient broth media.
2. 55°C incubator.

C. **STEPS:**

1. Inoculate steam autoclaved spore strip into liquid broth media.
2. Incubate at 55°C for seven days.
3. Observe every 12 hours for growth.

D. **RESULTS:**

1. Report any growth immediately to lab officer.
2. Report no growth at seven days.

TAB C-31

SODIUM AND POTASSIUM BY FLAME PHOTOMETER

A. **PURPOSE:** Blood is analyzed by flame photometer to determine the concentration of sodium and potassium.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Serum.
2. Controls.
3. Standards.
4. Lithium diluent.
5. Distilled water.
6. Autodilutor.
7. Flame photometer.
8. Operator's manual.

C. **STEPS:**

1. Start the flame photometer according to the operator's manual.
 - (a) Turn the propane screw valve completely open.
 - (b) After 10 seconds, turn the on/off switch to on. If the "flame on" indicator light does not come on within a few seconds, turn off the instrument.
2. Warm up the instrument for 5 minutes while aspirating the lithium diluent.
3. While aspirating the blank solution, zero the instrument. Using the sodium and potassium zero control knobs, set the sodium, and potassium digital displays to zero.
4. Standardize the instrument.
 - (a) Aspirate the 140/5 standard solution.
 - (b) Check the lithium response meter to insure that the red indicator is between the two lines.
 - (c) Using the sodium and potassium balance control knobs, set the sodium digital display to 140, and the potassium digital display to 5.0.
 - (d) Rerun the zeroing and standardizing procedures several times and make any necessary adjustments.
5. Quantitate the samples.
 - (a) Aspirate the control and unknown specimens in turn.
 - (b) With each aspiration, check the lithium response meter.

(c) With each aspiration, record the values from the sodium, and potassium digital displays.

NOTE: If the lithium response meter is outside the two lines at any time during your determinations, correct it with the lithium set control; then rezero and standardize the instrument before continuing the analysis.

6. Shut down the instrument.

(a) Aspirate a cleaning solution for at least 15 seconds.

(b) Aspirate deionized water to remove the cleaning solution from the line.

(c) Turn the propane screw valve completely closed.

(d) Watch for the "no gas" indicator light to come on and the "flame on" indicator light to go out.

(e) After the "flame on" indicator light goes out, turn the on/off switch to off.

7. Record and report the NA and K in mEq/L for the unknown and control specimens.

8. Refer to the operator's manual for the preventive maintenance schedule.

D. **RESULTS:**

1. Normal values.

(a) Na 135-148 mEq/l

(b) K 3.5-5.3 mEq/l

TAB C-32

CHLORIDE

A. **PURPOSE:** The combination of silver ions and chloride ions is a quantitative reaction that results in an insoluble precipitate of silver chloride. This reaction is carried out by passing a known, constant current between two silver electrodes immersed in an acid solution. The silver electrodes provide a constant generation of silver ions in a sample to form silver chloride. When all the chloride has been precipitated as silver chloride, free silver ions begin to appear, changing the solution conductivity. This change is detected by sensing electrodes and the titration time readout is stopped. The instrument displays this relative time in MMOLs (or MEQs) for chloride per liter.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Chloridometer.
2. Micropipette - 20 or 100 microliters.
3. Chloride diluent.
4. Chloride standard.
5. Controls.
6. Chloride reaction beakers.
7. Kim-wipes.
8. Wash bottle with deionizer water.
9. Electrode polish (silver polish).
10. Sample beakers.

C. **STEPS:**

1. Warm up instrument for 5 minutes.
2. Select sample size (20 or 100 microliters).
3. Condition the electrodes by adding acid-chloride solution to the fill line in a sample beaker.
4. Immerse electrodes by placing beaker on the platform and raising the platform.
5. Depress the condition button which starts the stirrer. (The readout will indicate that conditioning is in progress.)
6. When the stirrer stops, pipette an aliquot of 100 mmol/L chloride standard into the beaker.
7. Depress the titration button.
8. The standard should read 100 + OR - 2 if a 100 microliter volume is used (100 + or - 4 if 20 microliters is used).
9. If reading is outside specification check that pipette volume is correct for sample volume selected.

- (a) Repeat if sample volume is correct.
 - (b) If second reading is not within range re-calibrate the instrument.
10. To re-calibrate:
- (a) Repeat steps 2-8 using five titrations of 100 mmols/L chloride standard (at the 100 microliter sample volume).
 - (b) Calculate the mean of the five readings, which should be between 99.5 and 100.5 mmols chloride/L. If not adjust the 100 microliter control with a screwdriver and repeat titrations until the mean result is within the range 99.5 - 100.5.
 - (c) Select the 20 microliter volume and condition the electrodes again.
 - (d) Titrate 5 samples of 100 mmol/L chloride standard, and calculate the mean which should be between 99 and 101. (The 5 readings should be reproducible to within + or - 4.)
11. Once standard has been set, run controls, and unknowns:
- (a) Keeping electrodes immersed, pipette selected volume of sample into beaker.
 - (b) Depress titration button.
 - (c) Record results from readout display.
 - (d) Can continue titrating up to 20 samples.
 - (e) When "change reagents" is displayed, lower beaker, and empty out contents.
12. Rise with deionized water and dry with clean Kim-wipes.
13. When determinations are complete, remove beaker and allow the electrodes to dry naturally. (Do not leave electrodes immersed in reagents when instrument is not in use.)

D. **RESULTS:**

- 1. Error in pipetting techniques is the most common source of sample value error.
- 2. Know that iodine and bromine will combine with silver ions in the same manner as chloride and cannot be differentiated by chloridometer.
- 3. Reference range: 98-106 mmol/L (or meq/L).

TAB C-33

CO₂ DETERMINATION (BICARBONATE)

A. **PURPOSE:** The combination of carbon dioxide (as decarbonate and phosphoenol pyruvate are enzymatically converted to oxalacetate and phosphate in a reaction catalyzed by phosphoenol pyruvate carboxylase. The oxalacetate is then reduced to malate by malate dehydrogenase. This process reaction causes the oxidation of NaDH to NaD which decreases absorbance at 340 nm. This decrease in absorbance is proportional to the amount of carbon dioxide present in the sample.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Spectrophotometer.
2. Bicarbonate reagents.
3. Micropipettes - 10, 100, and 1000 microliters.
4. Pipettes - 5 and 10 milliliters.
5. Timer.
6. Test tubes.
7. Controls.
8. Test tube rack.
9. Decarbonate standards.

C. **STEPS:**

1. Make spectrophotometer settings according to manufacturer specifications.
2. Prepare reagents according to manufacturer instructions.
3. Zero the spectrophotometer with reconstitution solution.
4. Add 2 milliliters of reagents to test tubes. Mark one as the blank.
5. Add 10 microliters of standard, controls, and unknowns to their respective tubes. (Add nothing to Blank.)
6. After 5 minutes, but no later than 15 minutes, after the sample has been added, aspirate the reagent blank into the spectrophotometer and record the absorbance (A).
7. Aspirate and record the absorbance of the standard (B), and the unknown (C).
8. Calculations for CO₂ are:
$$\frac{A - C}{A - B} \times \text{conc. of STD}$$

D. **RESULTS:**

1. Icteric, hemolyzed, or lipemic specimens require a sample blank: add 100 microliters sample to 2.0 ml saline and read absorbance (D) at 340 nm. The adjusted carbon dioxide concentration is calculated using the following formula:
CO₂ mex/L = (A - D + C) x conc of std.

2. CO₂ reference range: 25 - 29 meq/L.

3. Sera with values 40 meq/L should be diluted one part sample with one part reconstitution solution and re-assayed. Multiply the result by two.

TAB C-34

GLUCOSE

A. **PURPOSE:** When glucose is phosphorylated with adenosine triphosphate in a reaction catalyzed by hexokinase, glucose-6-phosphate is produced. G-6-P is oxidized to 6 - phosphogluconate in a reaction catalyzed by glucose-6-phosphate dehydrogenase. During the reaction nad is reduced to nadh causing an increase in absorbance at 340 nm which is directly proportional to the amount of glucose in the sample.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Spectrophotometer.
2. Glucose reagent.
3. Pipettes - (1.0, 10, and 25 ml).
4. Micropipette - 10 microliters.
5. Timer.
6. Test tubes.
7. Glucose standards.

C. **STEPS:**

1. Make spectrophotometer settings according to manufacturer instructions.
2. Prepare reagents according to manufacturer directions.
3. Set spectrophotometer for adequate aspiration volume (approximately 1.0 ml).
4. Pipette 1.0 ml of reagent into appropriate test tube.
5. Add 10 microliters to the reagent blank test tube and mix.
6. At timed intervals add 10 microliters of 200 mg/dl standard and unknowns to their respective tubes and mix.
7. After five minutes aspirate the reagent blank into the spectrophotometer and record the absorbance (A).
8. Aspirate the standard, sample or control in the same manner, and record the absorbance.
9. Calculate glucose concentration using the following formula:

$$\text{MG/DL} = \frac{B - A}{C - A} \times \text{Concentration of standard}$$

Where:
"A" is the reagent blank
"B" is the unknown (control or patient)
"C" is the standard

D. **RESULTS:**

1. Results 60 MG/DL should be diluted one part sample with two parts

isotonic saline and re-assayed. Multiply result by three.

2. Normal range: 65-110 mg/dl.

TAB C-35

BLOOD UREA NITROGEN (BUN)

A. **PURPOSE:** The indirect method of measuring blood urea nitrogen utilizes the action of urease on urea to produce ammonia and carbonic acid. The ammonia released from the urease reaction is combined with α -ketoglutaric acid in the presence of glutamic dehydrogenase. In the process nadh is oxidized to nad causing a decrease in absorbance at 340 nm, and is proportional to the ammonium concentration which is directly proportional to the urea nitrogen concentration.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Spectrophotometer.
2. Bun reagents.
3. Bun standards.
4. Micropipette - 10 microliter.
5. Pipettes - 1, 5, 10, 25 ml.
6. Timer.
7. Test tubes.
8. Isotonic saline.

C. **STEPS:**

1. Make spectrophotometer settings according to manufacturer specifications.
2. Prepare reagents according to manufacturer instructions.
3. Set spectrophotometer for adequate aspiration volume and zero the instrument.
4. Pipette 1.0 ml of reagent into clean dry test tube.
5. Add 10 microliters of water standard, control, and unknown into respective tubes. Mix and quickly aspirate into cuvette.
6. Record absorbance at 15s (A_{15}) and read every 15s for 30s (A_{30}).
7. Calculate the change in absorbance at 30s by subtracting A_{15} from A_{30} . The other reading is used to check the linearity of the reaction rate.
8. If the change in absorbance is greater than 0.325 dilute one part sample with two parts isotonic saline and re-assay. Multiply result by three.
9. Calculate bun concentration using the following formula:

$$\text{Bun in mg/dl} = \frac{A_{UNK} - A_{BLK}}{A_{STD} - A_{BLK}} \times \text{concentration of std}$$

D. **RESULTS:**

1. Fluoride inhibits urease action.

2. Abnormal NH₃ levels give falsely elevated bun results.
3. Buns 90 mg/dl should be diluted 1 to 2 with isotonic saline and re-assayed. Multiply result by three.
4. Reference range: 7-18 mg/dl.

TAB C-36

CREATININE

A. **PURPOSE:** Creatinine in a protein-free supernatant of plasma or serum is reacted with alkaline picrate to form a color complex whose intensity is measured at 510 nm.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Spectrophotometer with a band width less than 20nm.
2. Test tubes (16x125).
3. Timer.
4. Graph paper.
5. Volumetric flasks 500 ml, 1.0 l.
6. Beakers, 500 ml.
7. Balance.
8. Graduated cylinder - 100 ml.
9. Hot plate.
10. Plastic bottle.
11. Pipettes: .5, 1.0, 2.0, 3.0, 5.0, 10.0 ml.
12. Reagents:
 - (a) Picric acid (reagent grade).
 - (b) Sodium hydroxide (reagent grade).
 - (c) Sodium tungstate dihydrate.
 - (d) Polyvinyl alcohol.
 - (e) Sulfuric acid (concentrated).
 - (f) Creatinine (reagent grade).
 - (g) Hydrochloric acid (0.1 mol/l).

C. **REAGENT PREPARATION:**

1. Picric acid (0.036 mol/l - Dissolve 9.16 g picric acid, in about 500 ml distilled water at 80 degrees centigrade. Cool and dilute to 1.0 l with dH₂O. Protect from light. Stable almost indefinitely.
2. Sodium hydroxide, 1.4 mol/l - Dissolve 54 g NaOH in about 500 ml of dH₂O. Cool and dilute to 1.0 l with dH₂O and store in a plastic bottle. Stable for 12 months at room temperature.
3. Tungstic acid, 0.035 mol/l. Dissolve 1.0 g polyvinyl alcohol with heat (do not boil) in 100 ml water, then cool and transfer to a 1.0 l volumetric flask

in which 11.1 g sodium tungstate dihydrate has been dissolved in approximately 300 ml water. In a separate container mix 2.1 ml concentrated sulfuric acid with 300 ml of water. Then mix the sodium tungstate and sulfuric acid solutions together and dilute the mixture to 1.0 l with water. (Mixture is stable at room temperature for at least 12 months. Do not refrigerate.)

4. Creatinine stock standard, 20 mmol/l - Dissolve 0.226 g creatinine in 100 ml of hcl (0.1 mol/l). This solution is stable at 4 - 6 degrees centigrade for 12 months. Prepare working standards volumetrically as follows:

<u>Stock standard (ml) diluted to 100 Ml with hcl, 0.1 mol/L</u>	<u>Plasma Concentration Equivalent mg/dl</u>
0.5	1.1
1.0	2.3
2.0	4.5
3.0	6.8
4.0	9.0
6.0	13.6
8.0	18.1

(Working standards are stable for at least one month.)

D. STEPS:

1. De-proteinize serum or plasma unknowns and controls by placing 4.5 ml tungstic acid in a 16 x 125 mm tube and adding 0.5 ml sample. Mix thoroughly for 10s and centrifuge at 1500 x g for 10 minutes.

2. Dilute urine samples 1:200 with dH₂O.

3. Pipet 3.0 ml de-proteinized supernatant, diluted urine or standards into the appropriate tubes. Also set up a reagent blank of 3.0 ml water.

4. Add 1.0 ml picric acid to each tube and mix thoroughly.

5. At accurately timed 30s intervals add 0.5 ml HaOH to each tube and mix thoroughly.

6. Exactly 15 minutes after adding haoh, read the absorbance of each tube at 500 nm against the reagent blank set to zero absorbance.

7. Plot absorbance versus concentration for standards and read concentration of unknowns and controls from the curve. Multiply concentration found for urine samples by 20; multiplying by 20 corrects for the original dilution of 1:200.

8. Use the same method for urine by diluting at 1:200 in HCl, 0.1 mol/l, in a tightly closed screw-capped tube, and placing the tube in a boiling water bath for one hour to form the concentrated product creatinine which is then determined by the method above on heated and un-.

9. Heated dilutions. Each of the two values obtained is multiplied by 20. The value of the unheated tube (preformed creatinine) is subtracted from the value of the heated tube (total creatinine), and the difference is multiplied by 1.16 to correct for the difference in creatinine and creatine molecular mass.

E. RESULTS:

1. Significant hemolysis of a blood specimen may cause spurious elevations of creatinine values for serum or plasma.

2. Reference ranges: male: 0.9 - 1.5 mg/dl*

female: 0.7 - 1.3 mg/dl

*Serum and plasma ranges only.

TAB C-37

AMYLASE

A. **PURPOSE:** In the presence of amylase, maltotetraose substrate is hydrolyzed to yield two moles of maltose per mole of substrate. Each mole of maltose is phosphorylated to a mole each of glucose and b-glucose-1-phosphate by the catalyzing enzyme maltosephosphorylase. B-phosphoglucomutase converts b-glucose-1-phosphate to glucose-6-phosphate which is then oxidized to 6-phosphogluconate as had is reduced to nadh in a reaction catalyzed by G-6-d. The production of nadh is measured at 340 nm and is directly proportional to the amylase concentration in the specimen.

B. EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:

1. Spectrophotometer.
2. Amylase reagent.
3. Pipettes - 40 microliters, 1.0, 5.0, and 10 ml.
4. Timer.
5. Test tubes.
6. Deionized (or distilled) water.

C. STEPS:

1. Make spectrophotometer settings according to manufacturer specifications.
2. Prepare reagents according to manufacturer instructions.
3. Set spectrophotometer for adequate aspiration volume.
4. Add distilled water to test tube. Aspirate the water into the spectrophotometer and adjust the absorbance to zero.
5. Add 1.0 ml aliquots of reconstituted reagent to an appropriate number of tubes.
6. At timed intervals add 40 microliters of samples or control to appropriate test tubes and mix thoroughly.
7. Aspirate samples. Record absorbance after three minutes (A_1 and read every 30s for two minutes (A_2).
8. Calculate the change in A/min by subtracting A_1 from A_2 and divide by two. The other readings were used to check linearity.
9. If the change in A/min is 0.145, dilute one part sample with two parts isotonic saline and re-assay. Multiply result by 3.
10. Calculate result using the following formula:

$$U/L = \frac{\text{Change in abs/min}}{a \times b \times v_s} \times 10^6 \times V_t$$

Where: 10^6 = Conversion of moles to millimoles
a = Molar absorptivity for nadh under these conditions (6.30×10^3 l/mole cm)

b = Light path of 1.0 cm (10 mm)
V_t = Total reaction volume (1.04 ml)
V_s = Sample volume (0.04)

Or in a simpler equation:

$$\text{A-Amylase (U/L)} = \text{Change in ABS/MIN} \times 4127$$

D. **REFERENCE RANGE:**

20 - 110 U/L.

TAB C-38

AST (SGOT)

A. **PURPOSE:** Aspartate aminotransferase catalyzes the transfer of an amino group from L-aspartate to 2-oxoglutarate resulting in the formation of oxalacetate and L-glutamate. Oxalacetate is formed which undergoes reduction with a simultaneous oxidation of nadh to nad in a malate dehydrogenase catalyzing reaction. Oxidation of NaDH causes a decrease in absorbance at 340 nm and the rate of change in absorbance is directly proportional to ast activity.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Spectrophotometer.
2. AST reagent.
3. Pipettes (100 microliters): 1.0, 5.0, 10.0, 25.0 ml.
4. Timer.
5. Test tubes.
6. Isotonic saline.

C. **STEPS:**

1. Make spectrophotometer settings according to manufacturer specifications.
2. Prepare reagents according to instructions.
3. Set sample time and vacuum for adequate aspiration volume (approximately 1.0 ml).
4. Pipette 1.0 ml of reagent into a clean dry test tube.
5. Add 100 microliters of control serum or patient sample into test tube. Mix and wait for one minute. Aspirate the mixture into the curette.
6. Record absorbance after 15x (A^{15}) and every 15s for one minute (A^{75}).
7. Calculate "Change in ABS/min" by subtracting A^{75} from A^{15} . Other readings are used as a check on linearity.
8. If the "Change in ABS/MIN" is 0.29, dilute one part sample with nine parts isotonic saline and re-assay. Multiply result by 10 to compensate for the dilution.
9. Calculate U/L by using the following formula:

$$\text{AST (U/L)} = \frac{\text{Change in ABS/MIN} \times 10^3 \times 1.1}{6.3 \times 1 \times 0.1}$$

$$= \text{"Change in ABS/MIN} \times 1736 \text{"}$$

Where :

6.3	= Millimolar absorptivity of NaHD
10^3	= Conversion of milliliter to liter
1	= Light path in cm
1.1	= Total reaction volume in ml
0.1	= Sample volume in ml

D. REFERENCE RANGES:

8-27 U/L.

TAB C-39

ALT (SGPT)

A. **PURPOSE:** Alanine aminotransferase catalyzes the transfer of an amino group from L-alanine to 2-oxoglutarate resulting in the formation of pyruvate and L-glutamate. Pyruvate is then reduced to lactate. As reaction occurs nadh is oxidized to nad in the lactate dehydrogenase catalyzed reaction. Oxidation of nadh causes a decrease in absorbance at 340 nm and the rate of absorbance is directly proportional to alt activity.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Spectrophotometer.
2. Alt reagent.
3. Pipettes - 100 microliters; 1.0, 5.0, 10.0, 25.0 ml.
4. Timer.
5. Test tubes.
6. Isotonic saline.

C. **STEPS:**

1. Make spectrophotometer settings according to manufacturer specifications.
2. Prepare reagents according to instructions.
3. Set sample time and vacuum for adequate aspiration volume (approximately 1.0 ml).
4. Pipette 1.0 ml of reagent into a clean, dry test tube.
5. Add 100 microliters of control serum or patient sample into test tube. Mix and wait for one minute.
6. Aspirate the mixture into the cuvette.
7. Record absorbance after 15s (A_{15}) and every 15s for one minute (A_{75}).
8. Calculate change in ABS/MIN by subtracting A_{75} from A_{15} . Other readings are used as a check on linearity.
9. If the change in ABS/MIN is 0.29, dilute one part sample with nine parts isotonic saline and re-assay. Multiply result by 10 to compensate for the dilution.
10. Calculate U/L by using the following formula:

$$\begin{aligned}\text{ALT (U/L)} &= \frac{\text{Change in ABS/MIN} \times 10^3 \times 1.1}{6.3 \times 1 \times 0.1} \\ &= \text{Change in ABS/MIN} \times 1746\end{aligned}$$

Where: 6.3 = Millimolar absorptivity of nadh
 10^3 = Conversion of milliliter to liter
1 = Light path in cm
1.1 = Total reaction volume in ml

0.1 = Sample volume in ml

D. REFERENCES RANGES:

Male 8-30 U/L

TAB C-40

BLOOD BANK CROSSMATCH LOG/ISSUING BLOOD

A. **PURPOSE:** To provide guidelines for accessioning specimens into the crossmatch log and for subsequently issuing blood for transfusion.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Green log book.
2. 3x5" index cards (patient ID and blood issue tag).

C. **STEPS:**

1. Accessioning.

Upon receipt of a specimen and request for blood or blood components:

(a) Verify that the specimen is correctly labeled with patient's register number, name, SSN, date, time, and drawers initials and the request is properly completed. Indicate the patient's name and SSN on a 3x5" index card, one for each unit requested. (IAW TAB C-1.)

(b) Assign the request the next sequential transfusion number (one for each SF 518).

(c) Annotate the transfusion number on the SF 518 and the 3x5" card.

(d) Indicate the expiration of the crossmatch on the 3x5" card.

2. Return two 3x5" card segments to the runner.

3. Log the request into the Crossmatch Log. The log contains the following information:

(a) Transfusion number (consecutive numbers beginning at 0001 - one number for each blood unit requested).

(b) Recipient's registration number.

(c) Recipient's name and SSN.

(d) Recipient's group and type.

(e) Donor unit number.

(f) Donor unit group and type.

(g) Donor unit expiration date.

(h) Compatibility results.

(i) Signature of person performing the crossmatch.

(j) Signature of the individual picking up the unit.

(k) Date and time unit is issued.

(l) Signature of individual issuing the unit.

(m) Disposition of unit.

4. Issue blood.

A 3x5" card will be presented to the Blood Bank when a unit of blood is needed for transfusion.

(a) Pull the unit with the same transfusion number from the refrigerator.

(b) Verify that the SF 518, and 3x5" card presented the ward representative and the crossmatch log all contain the same information.

(c) Check the color and appearance of the blood.

(d) Instruct the individual signing for the blood to verify that all information on the forms and the log book is correct.

(e) Instruct the individual to sign the log book for receipt.

5. Return of unused blood.

(a) Verify that the blood has not been at room temperature for longer than 30 minutes.

(b) Verify that the original transfusion request form is returned with the blood.

(c) Check the numbers on the SF 518, the bag, the tag, and the log book.

(d) Initial the log book and indicate the date and time the unit was returned.

(e) Witness that the person returning the blood initials the log book.

(f) Store blood in the refrigerator with other crossmatched blood or quarantine as necessary.

6. Return the empty blood bag.

(a) Ensure that Section III, record of transfusion on the SF 518 has been completed.

(b) Verify that no transfusion reaction was noted.

(c) Place empty blood bag in quarantine section of refrigerator for later destruction.

TAB C-41

FORWARD GROUP

A. PURPOSE:

1. Agglutination of red blood cells with a given antisera (Anti-A, Anti-B) indicates the presence of the corresponding antigen on the red cells and is a positive result. Absence of agglutination indicates the corresponding antigen is not present.

2. Anti-A, B will agglutinate red blood cells of blood groups A, B, and AB, but not group O. It is recommended for use along with Anti-A and Anti-B to provide an independent test confirming group O blood.

B. EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:

1. 10 x 75 mm tubes.
2. Pipettes.
3. Blood bank centrifuge.
4. Anti-A.
5. Anti-B.
6. Anti-AB.
7. 0.85% nacl.

C. STEPS:

1. To three tubes labeled A, B, and AB add one drop of the appropriate antisera.

2. To each tube add one drop of a fresh saline suspension (2-5%) of the cells to be tested.

3. Mix gently and centrifuge at the optimum speed and time for a saline test.

4. Gently re-suspend the cell button and examine macroscopically for agglutination.

5. Record graded results.

D. INTERPRETATION (REACTION WITH):

Anti-A	Anti-B	Anti-AB	Blood Group
=	=	=	O
+	=	+	A
=	+	+	B
+	+	+	AB
Agglutination		+	
No Agglutination	=		

TAB C-42

REVERSE GROUP

A. **PURPOSE:** Anti-A and Anti-B are naturally isoagglutinins which occur regularly in persons lacking the corresponding antigen. Testing against known A and B cells serves as a confirmation of the forward cell typing. As reagent cells will not routinely be available, cells from local donors may be substituted.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. 10 x 75 mm tubes.
2. Pipettes.
3. Immufuge.
4. A₁ Cells.
5. B Cells.

C. **STEPS:**

1. Re-suspend the reagent red cells evenly by gentle inversion. Do not shake.
2. Place two drops of the serum to be tested into each of the two tubes labeled "A" and "B."
3. Add one drop of A and B reagent cells to each appropriately labeled tube.
4. Mix gently and centrifuge at the optimum time and speed for the saline test.
5. Gently re-suspend the cell button and examine macroscopically for agglutination/hemolysis.
6. Record reactions.

D. **INTERPRETATION:**

Reaction with.

A ₁ Cells	B Cells	Blood Group
+	+	O
+	=	B
=	+	A
=	=	AB

+ = Agglutination or Hemolysis
= = No Agglutination

TAB C-43

Rh TYPING

A. **PURPOSE:** Agglutination of red blood cells with a given antisera (Anti-D) indicates the presence of the corresponding antigen on the red cells, and is a positive result. Absence of agglutination indicates the corresponding antigen is not present.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. 10 x 75 mm tubes.
2. 37° incubator.
3. Blood Bank centrifuge.
4. Anti-D.
5. Albumin.
6. 0.85% saline.

C. **STEPS:**

1. To a tube labeled "D" add 1-2 drops of Anti-D.
2. To a tube labeled "control," add 1-2 drops of Albumin.
3. To both tubes add one drop of a freshly prepared 2-5% saline suspension of the cells to be tested.
4. Mix well and centrifuge at the optimum speed and time for the albumin test.
5. Gently re-suspend the cell button and examine macroscopically for agglutination.
6. Record reactions.

D. **INTERPRETATION:**

1. Reactions with.

Anti-D		Rh Control	Rh Type
+	=		Rho (D) Positive
=	=		Rho (D) Negative - must be tested for the variant D ^u

2. If there is agglutination in the control tube the result of the Anti Rho(D) test is invalid and the cells must be tested with a reagent unaffected by protein abnormalities.

TAB C-44

D^u TEST

A. **PURPOSE:** Cells classified a D^u possess the Rho(D) antigen but are not agglutinated by some or all Anti-Rho(D) serums used. The antigen can be demonstrated by showing antibody attachment to the cell surface. Anti-globulin serum demonstrates whether or not this attachment has occurred after cells and anti-Rho(D) serum have been incubated together.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. 10 x 75 mm tubes.
2. 37°C incubator.
3. Pipettes.
4. Blood Bank centrifuge.
5. Anti-Rho(D).
6. Albumin.
7. 0.85% nacl.
8. Anti-human globulin.
9. Coombs control cells.

C. **STEPS:**

1. Continue with tubes from Rh typing.
2. Mix gently and incubate at 37°C for 20 minutes.
3. Wash cells in both tubes four times with large volumes of saline decanting completely after each wash.
4. Add 2 drops of anti-human globulin, mix gently, and centrifuge at the optimum speed and time for the Coombs test.
5. Gently re-suspend the cell button and examine macroscopically and microscopically for agglutination. Record the reactions.
6. Add one drop of Coombs control cells to all negative tubes, re-centrifuge, and re-examine macroscopically for agglutination. Record the reactions.

D. **INTERPRETATION:**

D ^u test	Rh control	Result
+	=	Rho(D) Positive
=	=	Rho(D) Negative
+	+	Not valid

Key: + = Agglutination
= = No Agglutination

E. **NOTES:**

1. If the autocontrol is positive, test results are not valid. The test should be performed using saline-agglutinating anti-Rho(D) for confirmation of Rho(D) type.

2. A Rho(D)_u negative individual with a positive direct Coombs will give a false positive D_u typing. In this case D_u status cannot be determined. Transfusion should be with Rh negative blood, if available.

3. Coomb's check cells must show agglutination. If no agglutination, the test is invalid and must be repeated.

TAB C-45

DIRECT ANTIGLOBULIN (COOMBS) TEST

A. **PURPOSE:** The direct antiglobulin test is used for the detection of invitro red blood cell sensitization. It is useful in the:

1. Diagnosis of hemolytic disease of the newborn.
2. Diagnosis of autoimmune hemolytic anemia.
3. Investigation of red blood cell sensitization caused by drugs.
4. Investigation of transfusion reactions.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Blood Bank centrifuge.
2. 10 x 75 tubes.
3. Disposable pipettes.
4. Patient's red cells -5% saline suspension prepared from EDTA sample.
5. 0.85% NaCl.
6. Coombs control cells.

C. **STEPS:**

1. Place one drop of a 5% saline suspension of the cells to be tested in a labeled 10 x 75 mm test tube.
2. Wash the cells three times with large volumes of saline.
3. Re-suspend in saline to make a 5% suspension.
4. Add 2 drops of Anti-human globulin (Coombs) and mix well.
5. Centrifuge at the optimum speed and time for the Coombs test.
6. Gently dislodge the cell button and examine for agglutination, macroscopically and microscopically.
7. Record graded results.
8. Add one drop Coombs control cells to all negative tubes.
9. Centrifuge at the optimum speed and time for the Coombs test and re-examine for agglutination (if the patient's cells were adequately washed in the first stage of the test, the control cells should be agglutinated, and the negative result on the patient is valid).
10. Record reactions.

D. **NOTES:**

1. Inadequate washing of the cells will result in the neutralization of the antiglobulin serum by trace amounts of globulin.
2. It is important that the antihuman serum be added immediately following completion of washing.

TAB C-46

TYPE AND CROSSMATCH

A. **PURPOSE:** To provide guidelines for the performance of a type and crossmatch procedure.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Tube of blood, red top tube.
2. 10 x 75 test tube.
3. 0.85% NaCl in squirt bottle.
4. Work card.
5. 37° water bath or heating block.
6. Anti A, Anti B, Anti AB, Anti D, albumin, anti-human globulin (Coombs Coombs Control Cells).
7. Blood Bank lamp (gooseneck with concave mirror).

C. **STEPS:**

1. Determine patient's abo group and Rh type as per those procedures.
2. Label all tubes with a unique identification for each patient (i.e., patient's initials).
3. Select the unit of blood according to the determined patient's group and type.
4. Cut a segment from each unit, and drain the blood into the tubes marked with donor number.
5. Prepare washed cell suspension of donor's blood by washing three times and suspending in a 3-5% suspension.
6. Mark a tube "auto control" and another tube for each unit of blood to be crossmatched with the unit number.
7. Place two drops of patient's serum into each tube.
8. Place one drop of patient's cells into the "auto control" tube.
9. Place one drop of donor's cells into the "crossmatched" tube.
10. Spin tubes for 15 seconds, gently re-suspend the cell button, and examine microscopically for agglutination or hemolysis. In an emergency situation, the blood may now be issued if no agglutination or hemolysis is observed. However, in all cases, the crossmatch is continued.
11. Add two drops of 22% bovine albumin.
12. Place all tubes in the 37° water bath or heating block for 15 minutes.
13. Centrifuge and read macroscopically.
14. Fill all tubes with 0.85% NaCl and wash three times, each time carefully blotting the edge of the tube.

15. Add two drops of anti-human globulin, spin and read microscopically.
16. Add one drop of Coombs control cells to all negative tubes, centrifuge, and read macroscopically. Record all reactions.
17. Blood units are ready for issue.
18. If incompatible:
 - (a) Recheck patient type.
 - (b) Recheck donor type.
 - (c) Check for cold agglutination.
 - (d) Crossmatch different units.
 - (e) Ask for assistance from supervisor.
19. Coombs control cells must show agglutination. If no agglutination, the test is invalid and must be repeated.

TAB C-47

EMERGENCY RELEASE OF BLOOD

A. **PURPOSE:** To provide guidelines for the release of blood in emergency situations.

B. **EMERGENCY - GROUP AND TYPE SPECIFIC BLOOD:**

1. In an emergency, it may be necessary to issue blood before completing the crossmatch. Blood of the same ABO group and Rh^o(D) type as the patient's may be so issued upon receipt of the following items:

(a) SF-518 marked "EMERGENCY" in the box labeled date and hour wanted, with an indication of the nature of the emergency in the "remarks" section.

(b) A properly labeled clot signed by a physician or an authorized nurse.

(c) Properly embossed 5x7" card.

2. Have the runner stand by while performing steps 3-5.

3. Enter in the sign out log transfusion number, date, patient's name, ward, and product ordered. Label the 5x7" card and SF-518 with a transfusion number.

4. Perform ABO cell and serum grouping and Rh^o(D) cell typing on patient's cells, according to methods already described.

5. Select blood unit from refrigerator. Perform ABO cell grouping and Rh^o(D) cell typing on the unit, using a suspension of unwashed cells from the segment.

6. Enter donor number and patient's and donor's ABO group and Rh^o(D) type, in SF-518. Written SF-518 "Group and Type Specific, no Crossmatch." Blood may then be issued to runner.

7. After issuing blood, proceed with complete crossmatch on the unit already issued, as well as other units requested. While crossmatch is incubating, complete patient data card and stamp it "Group and Type Specific, not Crossmatch" next to the appropriate unit number.

8. After 10 units of blood has been transfused, and no alloantibodies are present, a crossmatch is no longer required.

C. **EMERGENCY - O NEGATIVE PACKED CELLS:**

1. In extreme emergencies, even the above procedure may be too time consuming. In such instances a medical officer may ask for one or two units of group O, Rh^o(D) negative packed cells by personally notifying the Blood Bank. Note however, that O negative packed cells may be extremely limited and therefore should be issued with utmost consideration.

2. Upon receipt of such a request, O-negative packed cells may be issued at once to a responsible ward corpsman who brings a properly filled out SF-518 to the Blood Bank. Enter in the sign out log the patient's name, the date, and the transfusion number, and write in read O-negative packed cells no crossmatch on the Sign Out Log and SF-518.

3. Select unit of O-negative packed cells from refrigerator. Enter donor number in sign out log and on SF-518. Remove a segment to be used for

compatibility testing. Blood may then be issued to the runner.

4. After issuing blood, if a properly labeled clot tube has been received, proceed with complete crossmatch on units already issued, as described above. If not, return to the ward with the corpsman, obtain the clot, and proceed with the crossmatch.

TAB C-48

PREPARATION OF COOMB'S CONTROL CHECK CELLS

A. PURPOSE: To provide instructions for the preparation of Coomb's control check cells.

B. EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:

1. Ph-positive red cells.
2. Anti-Rh^o(D).
3. Saline.
4. 37°C water bath.

C. STEPS:

1. Obtain 1 ml of anticoagulated Rh-positive red cells and wash the cells three times with saline. (The red cells selected may be of any blood group.)
2. Suspend the Ph-positive red cells in saline to a 3% concentration.
3. Add to the suspension an equal volume of anti-RH^o(D), slide IgG serum, mix, and incubate in a 37°C water bath for 30 minutes (mix the suspension every 10 minutes during the incubation).
4. Centrifuge and remove the supernatant fluid (the cells should not agglutinate).
5. Wash the cells three times with saline and re-spin the cells to a 3% concentration with saline.
6. To test these cells, mix in equal parts with antiglobulin serum (the cells should not agglutinate).
7. Place the antibody coated cells in a labeled container with an expiration date of one week.

TAB C-49

DONOR SCREENING

A. **PURPOSE:** It is necessary to carefully screen blood donors in order to maintain maximum safety for both the donor and the recipient of blood products.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Donor cards.
2. Thermometer.
3. BP cuff.
4. Stethoscope.
5. Fingerstick items.

C. **STEPS:**

1. Donors will complete a Blood Donor Card. All sections except, Hct., temperature, pulse, and blood pressure will be filled out by the donor. The donor must also sign and date the card.

2. Lab personnel will determine the Hct., temperature, pulse, and blood pressure.

3. The following criteria for physical condition must be met:

- | | |
|----------------------|---|
| (a) Weight | greater than 110 lbs. |
| (b) Oral Temperature | less than 99.6°F (37.5°C) |
| (c) Pulse | regular and 50-100 beats/min. |
| (d) Blood Pressure | systolic 90-180 mm Hg.
diastolic 50-100 mm Hg. |

(e) Hgb. or Hct.

specific gravity (copper sulfate)

Female	greater than 1.053 (12.5 gm/dl Hgb)
Male	greater than 1.055 (13.5 gm/dl Hgb)

Hematocrit	Female	greater than 38%
	Male	greater than 41%

(f) Phlebotomy site must be acceptable, showing no rash, or recent wound.

4. Medical history.

(a) The medical history and donor card will be reviewed by a qualified member of the laboratory.

(b) Any item checked yes must be specifically reviewed by the screener. A notation for each item marked yes must be made on the back of the Donor Card with an explanation.

5. The following conditions will serve as a cause of permanent rejection:

- (a) Any history of viral hepatitis.
- (b) Donor who has had a confirmed positive for HBsAG or positive Western Blot.
- (c) Donor who has been or who is a drug addict.
- (d) If the donor gave a unit which was the only unit of blood or blood component or derivative administered to a recipient who within six months developed post-transfusion hepatitis, that donor will be permanently disqualified.

6. Temporary rejections.

- (a) Prospective donors shall be deferred for (12) months after the last injection of Hepatitis B Immune Globulin (HBIG).
- (b) Smallpox: deferred until after scab has fallen off or two weeks after an immune reaction.
- (c) Measles, mumps, yellow fever, oral polio vaccine, animal serum products: Deferred two weeks after last immunization.
- (d) German measles (Rubella) deferred two months after last injection.
- (e) Rabies (therapeutic): Deferred one year after last injection.
- (f) There must be at least an eight (8) week interval (no more than five (5) times per year) since the last blood donation by the donor or he/she is to be temporarily rejected until the interval is satisfied.

7. Donor will read information on AIDS and high risk groups for AIDS and will sign a statement which reads as follows:

"I have reviewed and understand the information provided to me regarding the spread of the AIDS virus by donated blood or plasma and if I consider myself to be a person at risk for spreading the virus known to cause AIDS, I agree not to donate blood or plasma for transfusion to another person or for further manufacture."

8. Donor will complete the self-exclusion form represented in TAB G-28.

D. **RESULTS:**

Exceptions may be warranted due to blood supply conditions. Exceptions must be approved by the Head, Laboratory Department.

TAB C-50

BLOOD DONOR PHLEBOTOMY

A. **PURPOSE:** To provide guidelines for the collection of blood from donors.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Labeled blood bag with labeled pilot tubes.
2. Donor prep kit.
3. Sterile gauze.

C. **STEPS:**

1. Identification.

Prior to starting phlebotomy:

(a) Identify donor record with the donor by name. Have the donor tell you his name and social security number.

(b) Check to be sure all numbered labels are the same on the donor card, the container, and the processing tubes.

(c) Processing tubes are for laboratory tests other than compatibility testing and must accompany the container during collection of blood. They may be attached in any convenient manner to assure correct identification.

(d) Double check all numbers.

2. Venipuncture site preparation.

(a) Select a large firm vein in an area that is free of skin lesions. It is often helpful to inspect both arms and to use a blood pressure cuff inflated to 50-60 mm Hg to make the veins more prominent. Having the donor open and close his hand a few times is also helpful. Release the pressure and prepare the site.

(b) Prepare a area at least 1 1/2 inches in all directions from the intended site of venipuncture. Use sterile materials and instruments or follow procedures in TAB H-2.

(c) Scrub area for 30 seconds with iodophor-pvp scrub, (with 0.75% titratable iodine). Excess foam must be removed but the arm need not be dry before proceeding to the next step.

(d) Apply iodophor-pvp, (with 1% titratable iodine) starting at site of venipuncture and moving outward in a concentric venipuncture.

(e) If not ready to do venipuncture immediately, cover the site with dry sterile gauze.

3. Phlebotomy and collection of samples.

(a) Inspect bag for any defects.

(b) Position bag carefully. Be sure counterbalance is level and adjusted for the amount to be drawn. Unless metal clips and a hand sealer are used, make

a very loose overhand knot in the tubing. Hang the bag and route the tubing through the pinch clamp.

(c) Reapply the blood pressure cuff. Have donor open and close hand until previously selected vein is again prominent.

(d) Uncover sterile needle and do venipuncture immediately. Tape the tubing to hold needle in place and cover site with sterile gauze.

(e) Open the temporary closure between the interior of the bag and the tubing following manufacture's instructions.

(f) Have donor open and close hand, squeezing a handgrip slowly and continuously during the collection.

(g) Mix the blood and anticoagulant gently and continuously during collection.

(h) Blood flow will stop after the proper amount has been collected.

(i) Bleeding should be completed fairly rapidly to prevent the triggering of the clotting mechanism. Units requiring more than eight minutes to draw may not be suitable for preparation of platelet concentrates or antihemophilic factor; however, if adequate blood flow is assured and constant agitation maintained, rigid time limits are not warranted.

(j) Seal the tubing 4 to 5 inches from the needle by making a white knot or using a metal clip.

(k) Grasp tubing on the donor side of the seal and press to remove blood for a distance of not more than an inch. Clamp with a hemostat.

(l) Cut tubing between seal and hemostat. To fill processing tubes for laboratory tests, other than compatibility testing, remove stopper from tubes, release hemostat, and allow blood to flow directly from vein. Re-identify tubes with container after filling.

(m) Deflate and remove blood pressure cuff. Remove needle from arm. Apply pressure and have donor raise arm (elbow straight) and hold gauze firmly over phlebotomy site with other hand.

(n) Discard needle assembly into special container designed to prevent accidental contamination to personnel.

(o) Starting at seal, strip donor tubing as completely as possible into the bag. It is important to work quickly before initiation of coagulation occurs.

(p) Invert bag several times to mix thoroughly; then allow tubing to refill with anticoagulated blood from bag. Strip tubing at least three times.

(q) Tubing must be left attached to the bag and sealed into sterile segments, suitable for compatibility testing, using knots, metal clips, or dielectric sealer. A final double seal should be made within 2 inches of the bag. It must be possible to separate from container without breaking sterility of container.

(r) Reinspect container for defects.

(s) Recheck numbers on container, processing tubes, and donor record.

(t) Place unit into refrigerator on the shelf labeled "Unprocessed Blood" until it is ready for further processing.

4. Care of the donor after phlebotomy.

(a) Check arm and apply elastoplast bandage after bleeding stops.

(b) When in satisfactory condition, allow donor to sit up. DO NOT LEAVE DONOR.

5. Donor should be given some simple instructions.

(a) Do not smoke for a half-hour.

(b) Eat and drink something before leaving.

(c) Do not leave until released by a staff member.

(d) Drink more fluids than usual in the next four hours.

(e) You may resume all normal activities after about a half-hour if you feel well.

(f) Remove the bandage after a few hours.

6. Send the donor over to the canteen area to get juice and something to eat.

D. DONOR REACTIONS:

1. Most donors tolerate donating very well, but occasionally an adverse reaction may occur. The reactions in order of increasing severity are:

(a) Feeling of faintness or giddiness.

(b) Vasovagal reaction (weakness, dizziness, diaphoresis, pallor, and nausea).

(c) Syncope.

(d) Convulsions.

2. For reactions b-d above, a physician should be consulted. The following are guidelines for treatment.

(a) At first sign of reaction, stop phlebotomy.

(b) Lower donor's head and elevate feet if possible.

(c) Loosen restrictive clothing.

(d) Be sure donor has adequate airway.

(e) Apply cold compresses to forehead.

(f) If nausea occurs, instruct donor to breathe slowly. Have emesis basin and tissues available.

(g) Notify physician immediately of convulsions.

(h) The nature and treatment of all reactions should be recorded on the donor card.

TAB C-51

PROCESSING DONOR BLOOD UNITS

A. **PURPOSE:** Each unit will have an ABO group, Rho(D) type, and atypical antibody screen. In this setting, it will not be possible to screen units for syphilis, hepatitis, HTLV-III, ALT, or anti-HBC.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

C. **STEPS:**

1. ABO group.

(a) Perform forward (red cell) group slide typing from a segment from the unit.

(b) Perform reverse (serum) group tube testing from one on the pilot tubes.

(c) Record results separately. Determine if discrepancies exist between forward and reverse grouping interpretations. DISCREPANCIES MUST BE RESOLVED PRIOR TO LABELING THE UNIT.

2. Rh_o(D) type.

(a) Perform Rh_o(D) slide typing from a segment from the unit. Include a Rh_o(D) control.

(b) Perform D_u testing (tube) on all Rh_o(D) negative units. If unit is D_u positive, it will be labeled with an Rh_o(D) positive label.

3. Atypical antibody screen: (Note: As reagent screen calls are not routinely available, this may not be possible to perform).

(a) Perform an atypical antibody screen using pooled reagent screen cells and serum from a pilot tube.

(b) The presence of atypical antibodies prevents the unit from being processed into fresh frozen plasma.

(c) The presence of an atypical antibody does not prevent the unit from being processed into packed red blood cells.

TAB C-52

BLOOD COMPONENT PREPARATION - RED BLOOD CELLS (HUMAN)

A. **PURPOSE:** To provide guidelines for the preparation of red blood cells (human).

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Dual beam balance.
2. Refrigerated centrifuge.
3. Rubber bands (as weights).
4. Plasma expressor.

C. **STEPS:**

Separation of plasma.

(a) Balance the unit using rubber weights, prior to placing into centrifuge. Use a dual-beam balance and bucket-style centrifuge heads.

(b) Place the rubber weights and units (base down) into centrifuge heads. Be sure to remove the plastic clip from the unit.

(c) Centrifuge in a refrigerated centrifuge for 5 minutes at 5,000 RPM (5,000 g).

(d) Place the primary bag containing centrifuged blood on the plasma expressor and release the spring, allowing the plate of the expressor to contact the bag.

(e) Penetrate the closure of the primary bag and allow the plasma to flow into the satellite bag.

(f) Apply a hemostat when the appropriate amount of supernatant plasma has entered the satellite bag.

(g) Seal the tubing between the blood collection bag and the satellite bag in two places.

(h) Make sure the satellite bag has the donor number attached and cut the tubing between the two seals.

2. Storage.

(a) Red blood cells (human) must be refrigerated at 1-6°C. Keep them on the shelf marked unprocessed blood until the processing is completed.

(b) The transfer pack containing the recovered plasma is placed in the recovered plasma box, or further processed into FFP.

TAB C-53

**BLOOD COMPONENT PREPARATION - SINGLE DONOR PLASMA (HUMAN),
FRESH FROZEN**

A. **PURPOSE:** To provide guidelines for the preparation of single donor plasma (human) fresh frozen.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Fresh plasma expressed into satellite bag (minimum of 225 ml).
2. Test tube.
3. Cardboard cartons.
4. Freezer -30°C)

C. **STEPS:**

1. Freeze, label side down to allow bubble to form.
2. When frozen, stand bag up.
3. If bubble moves, contents are not frozen.
4. Label the satellite bag with the following information: The label is filled out and inserted into the box which contains the frozen plasma. It is attached to the satellite bag after the plasma has been thawed, prior to issuing.

(a) Volume of plasma.

(b) ABO group and Rh type.

(c) Antibody screening result.

(d) Expiration date.

5. Storage:

(a) Fresh frozen plasma must be stored at -30°C or below. The expiration date is one year from date of collection.

(b) The unit must not be refrozen if thawing has occurred.

TAB C-54

THAWING FFP

A. **PURPOSE:** To provide guidelines for the thawing of single donor plasma (human) fresh frozen prior to issuing for transfusion.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Plastic centrifuge bag.
2. 37°C water bath.
3. Fresh frozen plasma.

C. **STEPS:**

1. Thawing should be at 37°C with frequent mixing. The FFP should be placed into a plastic centrifuge bag prior to thawing to prevent contamination by the water in the water bath. The plasma should be completely thawed and residual precipitate dissolved.

2. Thawed plasma must be infused as promptly as possible to prevent loss of activity of the short-lived blood coagulation factors. Units that have been thawed and not used must not be refrozen but can be converted to other use (placed into the recovered plasma box).

3. Unit must be given within six hours after thawing, if factor VIII is required. If factor VIII is not required, unit must be given within 24 hours. Store thawed FFP at 1 to 6°C.

TAB C-55

RECEIPT OF BLOOD SHIPMENT

A. **PURPOSE:** All units received into the Blood Bank will be processed as follows to ensure that they are in good condition and suitable for transfusion.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Centigrade thermometer.
2. Antiserum to confirm ABO/Rh.

C. **STEPS:**

1. Upon receipt of shipment, open box, and place a centigrade thermometer in between 2 units. Close lid for 5 minutes.

2. Read thermometer. Blood must be between 1 and 10°C. If not within the range, notify a lab officer.

3. Inspect each unit for color and appearance. Quarantine suspicious units.

4. Check inventory against DD 573 (Shipping Inventory of Blood Products).

5. Sign and date DD 573. Note any shipping discrepancies on the form.

6. Retain original copy of DD 573 in files. Return second carbon back to the shipper.

7. Confirm ABO/Rh on units.

TAB C-56

TRANSFUSION REACTION WORKUPS

A. **PURPOSE:** On occasion, the administration of blood or blood products may cause untoward effects, commonly called "transfusion reactions." Investigation, identification, correction and prevention are all part of the responsibility of the Blood Bank worker.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. 12x75 test tubes.
2. Centrifuge.
3. Blood Bank reagents in rack.
4. Glass marking pen.
5. Ink pen.
6. Pipets.
7. Saline bottle.
8. Blood sample.

C. **STEPS:**

1. Check the identification of the patient and the donor blood:

(a) Check the following to insure all identification matches.

- (1) SF 518.
- (2) Unit number.
- (3) Patient's arm band.
- (4) Patient data card.
- (5) Patient 3x5" card.

(b) If a mistake is found in the selection of the crossmatched blood, check to see where the proper unit is. It may have also gone to the wrong patient, thus two transfusion reactions.

2. Comparing the pre and post transfusion samples.

(a) Observe the pre and the post transfusion samples for the following:

- (1) Color of the serum.

a Pink or red discoloration present after the reaction but not before indicates the presence of free hemoglobin and destruction of red cells.

b In samples drawn 4 to 10 hours after transfusion, yellow or brown discoloration indicates the presence of increased bilirubin and other hemoglobin breakdown products.

3. Performing the Direct Antiglobulin Technique (DAT).

(a) If an antibody coats the incompatible transfused cells, without immediately destroying them, the DAT will be positive, with a mixed field appearance.

(b) Circulating antibody coated cells are rapidly removed, so if the sample were drawn several hours after the reaction, the DAT may be negative.

4. Interpretation of findings.

(a) In nearly all cases, negative findings indicate no hemolytic reaction has taken place.

(b) If the patients clinical condition strongly suggest a hemolytic reaction, further investigation is warranted despite the negative preliminary findings. A further investigation must be authorized by a laboratory officer.

TAB C-57

MORGUE ADMISSIONS

A. **PURPOSE:** To provide guidelines for the handling of morgue admissions.

B. **CRITERIA:**

A lab tech is on call at all times to assist in the admission of bodies to the morgue if necessary.

C. **STEPS:**

1. Ward personnel will prepare the body, identify the body with a toe tag, transport the body to the morgue, and notify the laboratory of the admission.

2. The ward person will log the body into the morgue log, and place the body in the morgue.

3. Decedent Affairs will direct the transportation of the bodies to other locations. Upon notification from Decedent Affairs, the lab tech will point out the bodies to be moved, log the body as released, and allow the body to be removed from the morgue.

TAB C-58

BLOOD COLLECTION - FINGER PUNCTURE

A. **PURPOSE:** To provide a method of obtaining blood samples from patients by finger puncture.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Sterile gauze pads (2x2).
2. 70% isopropyl alcohol.
3. Blood lancets.
4. Capillary tubes.
5. Bandages.

D. **CRITERIA:**

The finger puncture is used when a patient is burned severely or is bandaged so that the veins are either covered or inaccessible. It is also used when only a small amount of blood is needed.

E. **STEPS:**

1. Wash hands.
2. Assemble equipment.
3. Using the middle or ring finger, massage or "milk" the finger down toward the fingertip. Repeat this "milking" five or six times.
4. Cleanse the fingertip with alcohol and let dry.
5. Take the lancet and make a quick stab on the side of the finger (off-center). To obtain a large round drop, the puncture should be across the striations of the fingertip.
6. Wipe away the first drop of blood to avoid dilution with tissue fluid. Avoid squeezing the fingertip to accelerate bleeding as this tends to dilute the blood with excess tissue fluid, but gentle pressure some distance above the puncture site may be applied to obtain a free flow of blood.
7. When the required blood has been obtained, apply a pad of sterile gauze and instruct the patient to apply pressure, then apply a bandage.

TAB C-59

BLOOD COLLECTION - VENIPUNCTURE

A. **PURPOSE:** To provide a method of obtaining blood samples from patients by venipuncture.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Sterile gauze pads (2x2).
2. 70% isopropyl alcohol.
3. Tourniquet.
4. Vacutainer needles and holder.
5. Vacutainer tubes appropriate for the test to be performed.

D. **CRITERIA:**

1. Blood is obtained from arm veins.
2. When arm veins cannot be used due to bandages, IV fluid therapy, thrombosed, or hardened veins, the requesting physician will be notified.
3. Blood will not be drawn from an arm with IV fluids running into it.
4. Blood will be drawn from a patient sitting or lying.

E. **STEPS:**

1. Wash hands.
2. Assemble equipment.
3. Explain procedure to patient.
4. Apply tourniquet around arm with enough tension so that the vein is compressed but not the artery. A sphygmomanometer may be used instead of a tourniquet if a patient is difficult to draw. Inflate the cuff midway between systole and diastole.
5. Position patient's arm extended with little or no flexion at the elbow.
6. Locate a prominent vein by palpation. If the vein is difficult to find, it may be made more prominent by massaging the arm with an upward motion to force blood into the vein.
7. Cleanse puncture site with 70% alcohol and allow it to dry.

CAUTION: After cleaning the puncture site, only the sterile needle should be allowed to touch it.

8. "Fix" or hold the vein taut. This may be accomplished by placing the thumb directly under the puncture site and exerting a light downward pressure on the skin or placing the thumb to the side of the site and pulling the skin taut laterally.

9. Using a smooth continuous motion, introduce the needle into the side of

the vein at about a 15 degree angle with the skin (bevel of needle can either be up or down).

10. Holding the vacutainer barrel with one hand, push tube into the holder with the other hand and watch for flow of blood into the tube until filling is completed.

11. While holding the vacutainer with one hand, release tourniquet with the other.

12. Place sterile gauze over puncture site and remove needle with a quick, smooth motion.

13. Apply pressure to puncture site and instruct patient to keep the arm in a straight position. Have patient hold pressure for at least 3 minutes.

14. Take this time to invert any tubes that need to have anti-coagulant mixed with the blood, then label specimens.

15. Reinspect puncture site and apply bandage.

F. **RESPONSIBILITY:**

1. Lab technician is responsible for the safety of the patient while collecting blood samples.

2. Lab technician is responsible for correctly labeling blood samples.

TAB C-60

CLEAN CATCH URINE COLLECTION INSTRUCTIONS

A. **PURPOSE:** To provide instructions for the collection of clean catch urines.

B. **DEFINITION:** A clean catch urine is a sample that is collected so as to minimize contamination by skin flora or other external elements. This is the preferred specimen for routine urinalysis and urine culture.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Cotton balls.
2. Soap.
3. Urine container.

D. **CRITERIA:**

All patients will be instructed how to collect a clean catch urine when a routine urinalysis or urine culture is requested.

E. **STEPS:**

1. Males.
 - (a) Wash hands with soap and running water.
 - (b) Wash glans of penis with soap and water using cotton balls.
 - (c) Rinse the area thoroughly with cotton balls soaked in plain water.
 - (d) After passing a small amount of urine in the toilet, collect the urine specimen in the container provided.
 - (e) Label container with patient register number and name.
2. Females.
 - (a) Wash hands with soap and running water.
 - (b) Wash the labia with a downward motion, front to back, and the urethra opening with soap and water using cotton balls.
 - (c) Rinse the area thoroughly with cotton balls soaked with plain water.
 - (d) Spread the labia wide with finger tips.
 - (e) After passing a small amount of urine in the toilet, collect the urine in the container provided.
 - (f) Label container with patient register number and name.

TAB C-61

RECALL STAFF

- A. **PURPOSE:** To provide a system for the duty crew to recall on-call personnel.
- B. **DEFINITION:** N/A.
- C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**
Departmental Watch Bill.
- D. **CRITERIA:**
Additional staff is sufficiently augmented to meet increased patient load.
- E. **STEPS:**
 - 1. Senior technologist on duty will initiate recall when:
 - (a) Turn around time exceeds 1 hour.
 - (b) Directed by higher authority.
 - 2. The recall will be:
 - (a) Limited to the number of augmentees actively required.
 - (b) Reported to the Duty Laboratory Officer.
 - 3. On call personnel:
 - (a) Respond quickly as possible.
 - (b) Report to Senior Technologist.

TAB C-62

IMMEDIATE REACTION TO MEDICAL EMERGENCIES

- A. **PURPOSE:** To establish the protocol to react to medical emergencies.
- B. **DEFINITION:** Medical emergency is a situation causing a life threatening condition that requires immediate medical attention to sustain life.
- C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**
1. Equipment.
 - (a) Crash cart.
 - (b) Litter with blankets.
 2. Supplies.
 - (a) As provided on crash cart.
 - (b) As requested by attending physician.
 3. Forms.

Chronological Record of Patient Care (SF 600).
- D. **CRITERIA:**
- All equipment properly supplied and functional.
- E. **STEPS:**
- Shock.
- (a) Lay patient down with feet elevated.
 - (b) Keep patient warm.
 - (c) Notify medical officer.
2. Hemorrhage.
- (a) Apply direct pressure to area.
 - (b) Notify medical officer.
3. Pulmonary arrest.
- (a) Establish airway.
 - (b) Give mouth-to-mouth.
4. Cardiopulmonary arrest.
- (a) Establish airway.
 - (b) Start CPR.

- (c) Notify medical officer.
 - (d) Call code.
5. Obstructed airway.
- (a) Clear mouth.
 - (b) Four blows back, four ABD thrusts.
 - (c) Until airway opens.
 - (d) Notify medical officer.
6. Emergency procedure for adverse reaction to contrast agents.
- (a) With hives (urticaria), erythema, itching, or angioedema.
Notify attending physician.
 - (b) With the above and dyspnea (difficulty in breathing).
 - (1) Call for help immediately.
 - (2) Apply a tourniquet above the injection site to impede venous and lymphatic flow, but not arterial circulation.
 - (3) Protect airway, suction as needed.
 - (4) O2 high flow (10-15 L/min), by reservoir mask.
 - (5) Patient should be supine with legs elevated unless respiratory distress predominates.
 - a Assist the physician or nurse with the following:
 - 1 Start large bore IV with NS TKO.
 - 2 Epinephrine 0.5 mg 1:1000 SQ in opposite arm.
 - 3 Benadryl 50 mg IV push by physician.
 - b With BP less than 80 and patient critical.
 - 1 IV NS wide open.
 - 2 Epinephrine 1:10,000 0.2mg to 0.3 mg may be given very slowly IV push by physician.
 - 3 Benadryl 50 mg IV push by physician
 - c Transport to Casualty Receiving as soon as possible for further definitive care.
7. Simple fainting.
- (a) Lay patient down.
 - (b) Keep warm.
 - (c) Notify medical officer.

TAB C-63

MAINTENANCE OF LABORATORY GENERAL FILES

A. **PURPOSE:** To provide a system for maintaining laboratory general files.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES AND FORMS REQUIRED:** N/A.

D. **CRITERIA:**

1. Forms are filed in sequence - file number and chronological order.
2. Documents are easily retrievable.

E. **STEPS:**

1. The LCPO will:

(a) Assure all correspondence, message traffic and other files are maintained IAW SECNAVINST 5210.11C Standard Subject Identification Codes.

(b) Maintain any other file as directed by Head, Laboratory Department.

2. At a minimum, the file will contain:

(a) Departmental logs.

(b) Division officer records.

(c) Training records.

(d) Maintenance requests.

(e) Supply requests.

(f) Watch bills.

(g) Morbidity reports.

(h) Notices/instructions.

F. **RESPONSIBILITY:**

LCPO.

TAB C-64

LABORATORY MANUAL FOR WARD PERSONNEL

A. **PURPOSE:** This manual provides guidance and information to hospital staff personnel regarding laboratory services and requirements.

B. **INDEX:**

Section	Topic
C	Hours and Staffing
D	Information Required on Lab request forms
E	Specimen Labeling
F	Request Categories
G	Tests Offered
H	Critical Values
I	Specimen Rejection
J	Blood Bank Services/Requirements
K	Immunohematology Services/Requirements
L	Hematology Services/Requirements
M	Urinalysis Services/Requirements
N	Microbiology Services/Requirements
O	Chemistry Services/Requirements

C. **HOURS AND STAFFING:**

1. The laboratory operates seven days per week, twenty four hours per day.
2. The lab is staffed by two full crews, which work 0700-1900 and 1900-0700 respectively.
3. On Sundays, the laboratory operates with reduced staff. Only essential work is performed on that day.
4. Phlebotomy and other specimen collection is to be done by ward personnel. The laboratory has very limited capability in this regard.
5. For additional assistance, a laboratory supervisor is available on-board 24 hours a day.

D. **LABORATORY REQUEST FORMS:**

1. The minimum information required to be on laboratory request forms is listed below. The Laboratory may have to reject requests that are not filled out correctly to prevent compromising patient care.

- (a) Patient registration number (and name if known).
 - (b) Patient location (ward).
 - (c) Requesting physician.
 - (d) Time and date collected.
 - (e) Test requested: Each individual test box must be marked clearly or the test must be requested in legible writing.
 - (f) Priority (routine, ASAP or STAT).
2. Completed laboratory results must be periodically picked up by ward

personnel. Request forms are separated by ward and are located in the receiving area of the laboratory.

E. SPECIMEN LABELING:

1. All specimens submitted to the laboratory must have at least the following information securely affixed to the specimen:

(a) Patient's register number and name (first and last) if known.

(b) Specimens collected for blood product compatibility testing require additional information. See Blood Bank section of this manual.

2. Hepatitis (or other infectious disease): Please label both the specimen and the laboratory request for the protection of the staff.

3. Under no circumstances will a specimen be processed by the laboratory unless it is clearly labeled with the patient's identification.

F. REQUEST CATEGORIES:

1. Routine: This classification is applicable to the majority of test requests, and means that the specimens may be processed without haste or urgency. Routine tests will be performed within a reasonable period of time (generally within one working day). If the request is not marked, it will be considered routine.

2. ASAP (As Soon As Possible): These tests will be performed at the completion of tests in process (results generally available within 90 minutes).

3. STAT: The use of this term implies an immediate or urgent need for the test results for optimum patient care. At the top of the request form, please indicate a telephone extension at which the medical officer involved may be reached. All other work will be set aside until the emergency work is completed and the medical officer notified of the results.

Note: Unless special arrangements are made with a laboratory officer, only the tests listed on the STAT list, are available ASAP or STAT.

G. TESTS OFFERED: The following is a list of tests offered by this laboratory. Contact the laboratory officer for information regarding tests not appearing on this list.

<u>Test</u>	<u>Available STAT</u>	<u>SF Form</u>	<u>Tube</u>	<u>Reference Range</u>
CBC - WBC	Y	549	Lavender	4.8-10.8
HGB	Y	549	Lavender	14-18 (Male) 12-16 (Female)
HCT	Y	549	Lavender	42-52 (Male) 37-47 (Female)
DIFFERENTIAL	-	549	Lavender	
BODY FLUID CELL COUNT	-	555	N/A	Synovial less than 200 cells /cu mm Pleural less than 100 cells/cu mm
CSF CELL COUNT	Y	555	N/A	0-8 lymphocytes
PT	Y	549	Blue	10-13 seconds
PTT	Y	549	Blue	25-39 seconds
FIBRINOGEN	Y	549	Blue	150-400 mg/dl
MALARIA SCREEN	-	552	Lavender	None Seen

URINALYSIS

(ROUTINE & MICRO) Y		550	Urine	
PREGNANCY TEST	-	550	Urine	Negative
RPR	-	551	Red	NR
MONO TEST	Y	551	Red/Lav	Negative
GLUCOSE	Y	546	Red	65-110
BUN	Y	546	Red	7-18
NA (SODIUM)	Y	546	Red	135-148
K (POTASSIUM)	Y	546	Red	3.5-5.3
CL (CHLORIDE)	Y	546	Red	98-106
CO2 (CARBONDIOXIDE)	Y	546	Red	25-29
CREATININE	-	546	Red	0.9-1.5
				0.7-1.3(Female)
AMYLASE	-	546	Red	20-110
SGOT	-	546	Red	8-27
SGPT	-	546	Red	8-30
CULTURE & SENSITIVITY	-	553	N/A	N/A
BLOOD CULTURE	-	553	N/A	Negative
GRAM STAIN	Y	553	N/A	N/A
PARASITIC&FUNGAL EXAM	-	552	N/A	None Seen
OCCULT BLOOD	-	552	FECES	Negative
ABO/RH	-	556	Red	N/A
TYPE AND CROSS	Y	518	Red	Compatible
DAT	Y	556	Red	Negative

H. CRITICAL VALUES:

1. The following laboratory values are considered critical.

HEMATOCRIT	Less than 29%
HEMOGLOBIN	Less than 8 GM/DL
WBC	Less than 4,000 WBC/UL Greater than 20,000 WBC/UL
PT	Greater than 15 seconds (coumarin Greater than 30 seconds)
PTT	Greater than 45 seconds (Heparin Greater than 70 seconds)
SODIUM	Less than 125 MEQ/L Greater than 155 MEQ/L
POTASSIUM	Less than 3.0 MEQ/L Greater than 6.0 MEQ/L
BUN	Greater than 40 MG/DL
GLUCOSE	Less than 50 MG/DL Greater than 300 MG/DL
POSITIVE BLOOD CULTURES	
POSITIVE CSF CULTURES	
POSITIVE CSF STAINS	

2. When a critical value is determined, the laboratory technician will contact the ward by telephone (or in person).

3. The ward is then responsible for initiating appropriate action regarding the patient's care (i.e., notifying the medical officer).

I. SPECIMEN REJECTION:

1. Specimens submitted to the laboratory for testing may be rejected for several reasons. The major causes for rejection are as follows:

(a) Improper, incomplete, or illegible labeling of the specimens or requests.

(b) Specimens in containers which are grossly contaminated externally with the specimen (e.g., sputum, feces, blood).

(c) Improper specimens for test required. Examples include:

(1) Anticoagulated specimens for test requiring serum.

(2) Clotted specimens for tests requiring whole blood or plasma.

(3) Specimens which exhibit signs of delay in submission (e.g., clot retraction) for test requiring rapid processing.

(4) Hemolyzed or lipemic blood specimens for tests which are affected by hemolysis or lipemia.

(5) Insufficient blood in a tube in which the specimen /anticoagulant ratio is critical.

(6) Insufficient specimen of the test(s) requested.

(7) Specimens requiring special timing, patient preparation, handling, storage, or processing which are instead submitted in a random fashion.

(8) Specimens obtained from I.V. lines.

2. When specimens are received which must be rejected, an effort will be made to notify the requestor and to provide instruction for the proper submission of the specimen.

J. BLOOD BANK SERVICES/REQUIREMENTS:

1. Blood products stocked.

(a) Red blood cells (human).

(1) Composition: Total volume 225 to 350 ml containing 200 ml of packed red cells, supernatant plasma, and largely nonfunctional platelets and white cells. Minimum hematocrit: about 70%.

(2) Indications: Correction of inadequate hemoglobin-red cell mass.

(3) Availability: Routinely stocked.

(b) Fresh frozen plasma (FFP).

(1) Composition: 175 to 250 ml of plasma containing approximately 0.7 to 1.0 units of all coagulation factors per ml of fluid.

(2) Indications:

- a Correction of acquired and inherited coagulation deficiencies.
- b Replacement of fluid of plasma exchange.

(3) Availability: Routinely stocked. Preparation and thawing time is approximately 45 to 60 minutes. Thawed plasma must be used within 6 hours.

2. Definitions.

(a) STAT Implies an immediate life threatening situation due to loss of blood.

(b) ASAP Implies no immediate threat, but the use of blood is certain as soon as it is available.

(c) Routine Indicates that use of blood is not definite but that available blood is necessary, or that the use of blood is definite but can be predicted at some future time.

(d) Type and Crossmatch the specimen is ABO and RH typed. A suitable unit of blood is then tested against the patient's serum in such a manner as to detect incompatibilities. To perform a crossmatch on 2 units requires approximately one hour.

3. Specimens for type and cross may only be drawn by Medical/Dental/Nurse Corps Officers or laboratory staff.

4. Procedure for ordering blood or blood components. The following protocol must be followed to correctly order blood (or components) and to insure the safe transfusion of the blood (or components).

(a) For each unit of blood (or component) required, submit on completed SF 518.

(b) Positively identify the individual to be drawn.

(c) Draw a red top tube.

(d) Label tube with patient's registration number, patient's name, and SSN (if known), phlebotomist's initials, date and time drawn. A single large (15 ml) red top tube is adequate for up to six units of blood.

(e) Sign the SF 518 in the space marked "signature" (above "verification signature"), to indicate that you have drawn the blood and verified patient identity.

(f) Deliver specimen and SF 518's directly to the Blood Bank. A transfusion number will be assigned and a 3x5 card for each unit returned to the ward for inclusion in the patient's chart. This 3x5 card must be brought to the laboratory in order to pick up the unit for transfusion.

5. Emergency release blood.

(a) Request this only when it would be life threatening to wait for compatibility testing.

(b) If time permits, notify the laboratory as soon as an emergency

situation develops. This will minimize delays.

(c) Obtain a specimen, label it properly (especially important in emergency situations), and rush it to the laboratory.

(d) Group and type specific blood will be released in preference to O Negative blood. There is almost always time for the group and type of the patient to be determined. Therefore, the use of O Negative blood for all emergencies will not be available.

(e) The blood will still be crossmatched against the patient, even following its release. The physician will be immediately notified of any difficulty encountered.

(f) As soon as time permits, the proper paperwork must be submitted to the Blood Bank for completion. Likewise, this paperwork must be completed as to post transfusion data by the physician concerned.

6. Automatic release of crossmatched blood.

(a) All units of blood crossmatched on a particular day will be released at 1000 two calendar days later (e.g., blood crossmatched 1 May is released at 1000 on 3 May). Requests for an extension of the crossmatches must be brought to the attention of the Blood Bank and will be handled on a case by case basis. Generally, a crossmatch can be extended for 24 hours providing that the patient has not received any blood within the past 3 months and has not been pregnant within the past 3 months.

(b) Patient receiving multiple transfusions over a period of days will require frequent re-crossmatching. This must be accomplished within 48 hours of the expected time of the next transfusion. The Blood Bank will notify the ward when re-crossmatching is necessary and when available blood is released on these patients before the indicated automatic release date.

7. Issue of blood for transfusion.

(a) Only hospital personnel will pick up units for transfusion.

(b) Maximum number of units issued. Except for emergency situations, only one unit of blood will be issued for a patient at one time. Blood for only one patient may be picked up at a time.

(c) Blood issue tag (3x5 card). Blood/blood products will be issued to physicians and ward personnel upon presenting the 3x5 card to the Blood Bank. Sign out the unit. The completed transfusion form and unit will then be carried back to the ward.

(d) Do not request the blood until the physician is ready to use it. If the transfusion cannot be started immediately, the blood product must be returned to the Blood Bank.

(e) Storage of blood. Safety considerations prohibit the storage of blood in any area except in the Blood Bank. Any blood maintained on the ward for more than 30 minutes and not used must be discarded by the Blood Bank.

8. Transfusion procedures.

(a) Who may start a transfusion. Only a medical officer, an I.V. certified registered nurse, or a nurse anesthetist may start a transfusion. Ultimate responsibility for the proper compliance with transfusion regulations

rests with the physician who ordered the transfusion.

(b) Patient identification. The attending nurse will identify the patient by checking the patient's identification band against the SF 518 and against the copy of the SF 518 attached to the unit.

(c) The transfusionist will complete the bottom left portion of the SF 518 and sign it. Another person will independently verify all the identification information and will sign the "verified by" block (bottom, left portion of SF 518). The transfusion may now begin.

9. Infusion set. All blood and blood products must be administered through a standard blood infusion set containing the appropriate filter. Blood and blood products must not be mixed with any intravenous solutions or medications, with the exception that packed red blood cells may be mixed with normal saline to facilitate infusion. All tubing containing blood must be rinsed with normal saline before being used for other agents. Certain I.V. solutions may be directly hemolytic in vitro.

10. Rate of infusion. During the first fifteen minutes, administer the infusion rather slowly with close supervision of the patient. If no adverse reaction is observed, the blood or blood product may then be administered as quickly as possible without provoking problems with fluid overload. The time required for completion of the infusion must not exceed 4 hours per unit.

11. Post-transfusion procedures.

(a) Following completion of the transfusion the attending nurse will complete and sign the "post transfusion data" portion of the SF 518 (bottom, right side).

(b) Place the top original white copy of the SF 518 into the patient's chart for a permanent record of the transfusion.

(c) Return the bottom copy of the SF 518 and the empty blood bag to the laboratory. Do not discard the blood bags on the ward.

12. Transfusion reactions.

(a) If a transfusion reaction is suspected, discontinue the transfusion immediately!!! Allow the blood product to remain hanging and keep the vein open by infusion of normal saline.

(b) Immediately notify the attending physician and the Blood Bank.

(c) The pathologist will be notified by the Blood Bank and will consult with the patient's physician to determine the nature and severity of the problem and the diagnostic tests which will be performed to evaluate the reaction.

(d) If the pathologist requests that a workup be performed, he will indicate what samples need to be submitted.

K. **IMMUNOHEMATOLOGY SERVICES/REQUIREMENTS:**

1. Specimens for immunohematology studies.

a. Except as stated, all specimens will be clotted whole blood, carefully drawn so as to avoid gross hemolysis. It is preferable to submit a 15 ml clot tube (red top) whenever possible.

b. SF 550 (immunohematology) or SF 557 (miscellaneous).

2. Blood group and type.

(a) Specimen: One red top tube.

(b) ABO group and RH type.

(c) Discussion: Blood types are not routinely returned to the ward when a type and crossmatch/type and screen is performed (unless the blood is actually used); submit an SF 550 along with the transfusion form, if the results of the blood typing are needed.

(d) Du: The designation RH negative Du positive is not used by this laboratory. Patients who are Du positive are RH positive and will be reported as such. Questions may be directed to a laboratory officer.

L. **HEMATOLOGY SERVICES:**

1. A CBC consists of a WBC, hgb and HCT.

2. A differential includes WBC differential, a platelet estimate, and RBC morphology. Differentials are automatically performed if the WBC count is less than 4,000 or greater than 11,000. Within this range, a differential will be performed only if specifically requested by the physician.

3. From a full 7 ml lavender top tube (EDTA), a CBC, differential and mono test can be performed.

(a) The tube must be full and have no clots.

(b) Completely mix the blood with the anticoagulant immediately after drawing to prevent clotting.

4. Coagulation testing (prothrombin time, partial thromboplastin time, fibrinogen level) requires a full 5 ml blue top (citrate) tube. The tube must be full since the ratio of anticoagulant to blood is critical for coagulation studies. Short draws, clots, or hemolysis are criteria for rejection of the specimen. Specimens over an hour old will not be accepted for analysis, since coagulation factors degrade very rapidly. NOTE: Do not overfill the blue top tube. The vacuum seal will allow just the right amount to be drawn.

M. **URINALYSIS SERVICES/REQUIREMENTS:**

1. Specimen submission.

(a) The first morning urine (most concentrated) is the specimen of choice. The specimen should be a fresh, clean voided urine.

(b) All urine specimens must be delivered to the laboratory within two hours of collection.

2. Routine urinalysis.

(a) Routine chemical urinalysis is performed with a multiple chemistry reagent strip and includes ph, protein, glucose, ketones, bilirubin, blood, nitrite, urobilinogen, and ascorbic acid.

(b) Microscopic examination will only be performed on urines with a positive chemistry, or upon specific request.

3. Specific gravity is determined using a refractometer.

N. **CHEMISTRY SERVICES/REQUIREMENTS:**

1. Testing offered: Profile or panel testing is not performed in this laboratory. All tests must be requested individually by marking the box next to the desired test or indicating the test in writing. Each test is performed individually in this laboratory. Please request only those tests needed.

2. Fasting requirements: For most routine chemistries, a fasting specimen is the ideal specimen. Eating prior to blood collection may lead to erroneous results, depending on the test. Tests directly affected by eating glucose. Lipemic serum resulting from a fatty meal may interfere with a number of tests. Therefore, whenever possible, a fasting specimen should be submitted for routine chemistry testing. Hemolysis may also alter test results. If present, hemolysis will be noted on the request form.

3. Turn around time: Turn around time varies with the workload, test requested, and urgency of request. In general, results can be expected as follows:

- | | |
|-------------|---------------------------------|
| (a) STAT | 1-2 hours |
| (b) ASAP | 2-4 hours |
| (c) ROUTINE | Most on day following the order |

TAB C-65

STANDARDIZED ARMED SERVICES BLOOD PROGRAM REPORTS

A. **PURPOSE:** To provide instructions for the submission of wartime reports. These instructions are applicable to all UNICOM Blood Program activities.

B. **STEPS:**

1. A Blood Report (BLDREP) and Blood Shipment Report (BLDSHIPREP) are the two standardized reports to be used in the worldwide Armed Services Blood Program. The reports will be used for transmission of blood status via various communication modes to include the KL-43/43A Off-line Electronic crypto device. The Blood Report/Blood Shipment report menu (BRM) will be used to prepare each report.

2. The respective Joint Blood Program Office may assign codes for the individual component blood program activities. Military map coordinates will be used for activity location. Location will be reported on first report and upon relocation. Blood requests from naval vessels should contain a projected location in order to coordinate delivery of blood products.

3. Blood requests should normally be based on a random distribution. Group and type specific blood should be transfused unless not available or medically contraindicated. Certain designated medical treatment facilities (MTF) will require group O blood only. Upon activation, each MTF should request a base load of blood products.

4. Blood shipments will normally be red blood cells. Blood group and type distribution from CONUS blood donor centers will be as follows: O POS (40%); A POS (35%); B POS (8%); O NEG (10%); A NEG (5%); B NEG (2%). Group AB blood will not be shipped.

5. Blood report messages should be minimally classified. Information copies should be kept to a minimum and specifically required by the respective OPLAN. Messages will be sent as immediate due to very short blood expiration dates. Correct plain language addresses from a current directory must be used.

6. Blood program facilities identification will be abbreviated by alphanumeric characters followed by the facility type code from the BRM. Example: 609CONT-H (609th Contingency Hospital - Medical Treatment Facility).

7. As requested, codes from the Blood Report Menu will be in the following order of modifiers: management, facilities, amount. Example: 32CSH-H200JRX25MVX/.

C. **SPECIAL INSTRUCTIONS:**

1. Medical treatment facilities (MTF). Each MTF will submit a blood report to a supporting blood supply unit as required.

2. Blood supply unit (BSU). The BSU blood manager will submit a blood report to an Area Joint Blood Program Office on the status of blood products in the BSU as required.

3. Area Joint Blood Program Office (AJBPO). The AJBPO will submit a blood report to the Joint Blood Program Office on the status of blood products in the BTCs and BSUs as required.

4. Joint Blood Program Office (JBPO). The JBPO will submit a Blood Report to the Armed Services Blood Program Office on the status of blood products in each of the Joint Blood Program areas.

BLOOD REPORT/BLOOD SHIPMENT REPORT

MENU

MODIFIERS:

MANAGEMENT: A. Joint Blood Program Office (JBPO)
B. Area Joint Blood Program Office (AJBPO)
FACILITIES: C. Armed Services Whole Blood (ASWBPL)
Processing Laboratory
D. Blood Donor Center (BDC)
E. Blood Products Depot (BPD)
F. Blood Transshipment Center (BTC)
G. Blood Supply Unit (BSU)
H. Medical Treatment Facility (MTF)
I. Naval Vessel (NV)

PRODUCTS: J. Red Blood Cells (RBC)
K. Whole Blood (WB)
L. Frozen Red Blood Cells (FRBC)
M. Fresh Frozen Plasma (FFP)
N. Frozen Platelets (FP)

BLOOD GROUPS: Q. Random Group and Type O,A,B
R. Random Group and Type O,A
S. Random Group and Type O
T. Random Group and Type A
U. Random Group and Type B
V. Random Group and Type AB

TIME FRAME: W. Required within 12 hours
X. Required within 24 hours
Y. Required within 48 hours

MISCELLANEOUS: Z. Not applicable or see remarks

BLOOD REPORT

FORMAT

LINE ONE Reporting unit name or code, type activity, and location.

LINE TWO Total number of blood products by group and type at end of report period.

LINE THREE Total number of blood products by group and type requested and time frame needed.

LINE FOUR Estimate of total number of blood products by group and type to expire next 7 days.

LINE FIVE Estimate of total number of blood products by group and type re-supply requirements next 7 days.

LINE SIX Narrative. Free text for comments and additional operational reporting requirements.

BLOOD SHIPMENT REPORT

FORMAT

LINE ONE Reporting unit name or code, type activity, and location.

LINE TWO Total number of each blood product by group and type in shipment. In following order: O POS/A POS/B POS/AB POS/O NEG/A NEG/B NEG/AB NEG//TOTAL//

LINE THREE Air bill or transportation control number: (TCN).

LINE FOUR Airline and flight number or Military Air lift Command (MAC) mission number at destination.

LINE FIVE Estimated date, time, group of shipment arrival at destination.

LINE SIX Number of boxes in shipment.

LINE SEVEN Point of contact and twenty-four hour telephone number.

LINE EIGHT Narrative. Open text for comments.

SAMPLE MESSAGE

FM: MEDICAL TREATMENT FACILITY

TO: BLOOD SUPPLY UNIT

INFO: AS DETERMINED BY COMMAND OPLAN

CLASSIFICATION

SUBJ: BLDRPT (AS OF DTG)

1. 32CSH-H
2. 200JR50MV
3. 150JRX25MVX
4. 25JS5JT
5. 1400JR
6. EXP 10J/RECD 50JR 24AUG86/REFRIG NEEDS REPAIR

SAMPLE MESSAGE

FM: BLOOD SUPPLY UNIT

TO: AREA JOINT BLOOD PROGRAM OFFICE

INFO: AS DETERMINED BY COMMAND OPLAN

CLASSIFICATION

SUBJ: BLDRPT (AS OF DTG)

1. 655-G
2. 2000JR
3. 2000JQY200 MVY
4. Z
5. 1400JQ
6. RECD 1000JR 23AUG86/ISSUE 32CSH50JR/

SAMPLE MESSAGE

FM: BLOOD TRANSHIPMENT CENTER

TO: AREA JOINT BLOOD PROGRAM OFFICE

INFO: AS DETERMINED BY COMMAND OPLAN

CLASSIFICATION

SUBJ: BLDRPT (AS OF DTG)

1. LH-F
2. 6000JQ
3. Z
4. Z
5. Z
6. RECD FM ASWBPL1 3600JQ/ISSUE 655 1000JR

SAMPLE MESSAGE

FM: AREA JOINT BLOOD PROGRAM OFFICE

TO: JOINT BLOOD PROGRAM OFFICE

INFO: AS DETERMINED BY COMMAND OPLAN

CLASSIFICATION

SUBJ: BLDRPT (AS OF DTG)

1. A1-B
2. 655-G 2000JR/ROTA-G1500JR/ZWEI-G 1000JR25MV
3. 655-G 2000JQY200MVY/ROTA-G 200JQY
4. Z
5. 40000 JQ
6. EXPECT FOUR USA HOSP TO ACTIVATE NEXT 2 DAYS.

SAMPLE MESSAGE

FM: JOINT BLOOD PROGRAM OFFICE

TO: ARMED SERVICES BLOOD PROGRAM OFFICE

INFO: AS DETERMINED BY COMMAND OPLAN

CLASSIFICATION

SUBJ: BLDRPT (AS OF DTG)

1. J1-A
2. A1-B 4500JR 25MV/A2-B2500JR/A3-B1700JR
3. LH-F 3600JQ/TORR-F 3600JQ
4. Z
5. 70000JR
6. EXP BLOOD REQ IAW OPLAN NEXT TWO WEEKS.

SAMPLE MESSAGE

FM: ARMED SERVICES WHOLE BLOOD PROCESSING LABORATORY

TO: BLOOD TRANSSHIPMENT CENTER

INFO: AS DETERMINED BY COMMAND OPLAN

CLASSIFICATION

SUBJ: BLDSHIPREP (AS OF DTG)

1. ASWBP1-A MCGUIRE AFB NJ
2. J/35/32/7/0/9/5/0/0//88
3. AB 11213
4. MAC 767
5. 211400Z MAR 86
6. 4
7. LT J. READY AV 383-6614/6615
8. BLOOD ICED 210600 MAR 86

D. **BLOOD PLANNING FACTORS:**

1. Blood planning factors are contained in ref A. These factors will be used by all component blood program activities.

2. Red blood cell group and type distribution will be as listed below. Group AB red blood cells will not be shipped.

BLOOD GROUP/TYPE	DISTRIBUTION
O Rh Positive	40%
A Rh Positive	35%
B Rh Positive	8%
O Rh Negative	10%
A Rh Negative	5%
B Rh Negative	2%

3. Sample calculation to be used for blood requests for 100 admissions. Designated combat zone medical treatment facilities may be directed to use only group and type O blood.

BLOOD GROUP/TYPE	CALCULATIONS	UNITS NEEDED
O Rh Positive	100x.40x2	80
A Rh Positive	100x.35x2	70
B Rh Positive	100x.08x2	16
O Rh Negative	100x.10x2	20
A Rh Negative	100x.05x2	10
B Rh Negative	100x.02x2	4
	TOTAL	200

E. **NOTES:**

1. Each MTF which includes naval vessels, submits a blood report to a designated component Blood Supply Unit (BSU).

2. Each designated component Blood Supply Unit issues blood products to MTF.

3. Each component Blood Supply Unit submits a daily blood report to a designated AJBPO. Information copies provided IAW component OPLAN.

4. Each Blood Transshipment Center submits a daily blood report to a designated AJBPO.

5. Each BTC submits a daily blood report to a designated AJBPO.

6. AJBPO cross-levels blood products between component BSU and submits daily blood report to JBPO. Information copies provided to each component Command Surgeon Office.

7. JBPO cross-levels blood products between blood program areas and submits daily blood report to ASBPO. Information copies provided to each AJBPO.

8. ASBPO directs designated ASWBPL to ship blood products.

9. ASWBPL ships blood products to designated theater Blood Transshipment Center.

10. CONUS Military BDCs ship blood products to designated ASWBPLs. Contracts with civilian blood agencies are activated when military blood program shortfalls are experienced.

11. Pre-positioned frozen blood products stored in theater BPO will be issued to BTCs and/or Blood Supply Units as directed by JBPO.

TAB C-66

DISPOSAL OF CONTAMINATED ITEMS

A. **PURPOSE:** To provide guidelines for the disposal of needles and contaminated waste.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Bleach.
2. Disinfectant solution.

C. **STEPS:**

1. Cleaning vacutainer holders.

Make up a solution of 1 part water, and 1 part bleach. Soak vacutainer holders for several hours or until the end of the day. Rinse and dry them on a paper towel. Change the bleach solution when needed.

2. Needle disposal.

(a) Dispose of needles in a container marked "needles for disposal." The container should be a thick cardboard box with all ends bound with tape to prevent needles from protruding.

(b) When the container is full, incinerate, and dispose as trash.

3. Contaminated specimens.

(a) All specimens received by the laboratory are to be considered hazardous and contaminated. However, specimens that have been identified as infectious should be handled with extreme care as outlined.

(b) If blood is collected from patients in isolation, all routine hospital procedures for isolation will be followed as established by the Nursing Service.

(c) Contaminated specimens that are spilled on the deck or counter tops are to be cleaned up with a suitable germicidal cleaner.

(d) Contaminated specimens will be disposed of by incineration, then disposed as trash.

(e) If contaminated material is pipeted into the mouth or penetrates a break in the skin, immediately flush with water and report to sick call for treatment.

TAB C-67

QUALITY CONTROL

A. **PURPOSE:** To describe the basic guidelines for a successful quality control program.

B. **CRITERIA:**

1. Quality control encompasses all laboratory areas from specimen collection and personnel to the finished laboratory report.

2. Quality control begins with qualified personnel. Their education and professional qualifications should meet the job requirements. Job descriptions must be available for each position; they must enumerate qualifications needed, duties, and responsibilities. Continuing training for personnel is essential.

3. Specimen collection must be controlled so that an adequate sample, properly identified by an affixed label, arrives in the laboratory. The specimen must be so collected that the patient is not traumatized and the specimen is not hemolyzed. All personnel drawing blood must be properly trained to avoid errors caused by such things as the tourniquet being left on too long. Persons collecting capillary samples must be made aware that the first drop of blood should be wiped away because it contains tissue juices. Other sources of error are cold and cyanotic skin and vigorous squeezing after the puncture.

4. Anticoagulants must be chosen to meet the requirements of the test. Consideration should be given to the stability of the specimen and effects on specific test requested.

5. Procedures should be selected from well accepted methods. New methods being considered should be compared with the existing methods and preferably with reference methods where available. Once a method is chosen, it then becomes necessary to define the normal ranges.

6. Controls must be run on a scheduled basis for all tests in the laboratory. Charts indicating these results should be displayed. Controls are generally run each time the test is done to determine if the equipment and reagents are functioning properly. Duplicate samples are run throughout the day to determine precision. Reference solutions must be run daily to determine accuracy.

7. The laboratory must have logbooks of all specimens received. They should be listed by patient identification number, with results of all tests retained. The log book should also contain the date, tests ordered for each patient and a designation of the patient's location. Record keeping must be organized and must include sufficient checks to keep clerical errors to a minimum.

8. A good procedure manual for each department is essential to the proper operation of the laboratory. Each procedure in the manual should be periodically reviewed for accuracy and clarity.

9. The final laboratory report must be in such a form that the physician can use it. It must identify the patient, give the patient's location, and the date and time the sample was collected. Significant results should be marked to bring them to the physician's attention.

10. There must be an established schedule of periodic preventive maintenance of all instruments used in the laboratory and proper records must be kept of all maintenance performed on each instrument. The methods of checking and testing must be defined for each piece of equipment. Revolutions per minute

requirements, timers, must be monitored at least once per month. Baseline or background checks must be performed each day of use for those instruments requiring such monitoring. Automatic diluters and samplers must be checked for accuracy at least once a month. Refrigerators and freezers in which reagents or specimens are stored must be set for the proper temperature and be capable of maintaining a temperature within $\pm 4^{\circ}\text{C}$ of that setting; they must be monitored at least once per day, and the results recorded. Incubators must be set for the specified temperature and must be capable of maintaining temperatures within $\pm 1^{\circ}\text{C}$ of that setting; they must be monitored at least once a day, and the results recorded. Water bath and heating block temperatures must be confirmed each day of use by thermometer check. All volumetric glassware, including pipettes and manual pipettors, should be checked for calibration before being put into routine use.

TAB C-68

GLASSWARE CLEANING

A. **PURPOSE:** To provide guidelines for the cleaning of laboratory glassware.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Alconox, prepared with hot water.
2. 5% HCL (v/v).
3. Pipette washer.

C. **STEPS - Routine glassware:**

1. Soak glassware for 2-4 hours in dilute alconox or 2% v/v stripping detergent prepared with hot water.
2. Rise thoroughly with tap water.
3. Soak in 5% v/v HCL acid solution.
4. Rinse with tap water.
5. Rinse with distilled water, drain, and dry in oven.

D. **STEPS - Pipettes:**

1. Immediately after use, place pipettes tip up in the pipette holder, with alconox detergent.
2. Using the pipette washer, place the carrier in the washer and turn on the water supply. Regulate the water supply until water circulates 12-15 times per hour.
3. After two hours, remove the carrier and allow water to drain. Place the carrier in a container of distilled water until the pipettes fill, lift the carrier, drain, and fill pipettes again.
4. Go through three changes of distilled water for adequate rinsing.
5. After the final rinse, allow pipettes to drain for 10 minutes and place in hot oven to dry.

TAB C-69

STERILIZER OPERATION

A. **PURPOSE:** To provide guidelines for operation of the field sterilizer.

B. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Demineralized water.
2. Sterilizer.

C. **STEPS:**

1. With control valve in the off position, energize unit.
2. Allow jacket pressure to reach the pressure of 27 psi.
3. Fill chamber with proper amount of water.
4. Load sterilizer.
5. Set timer as required.
6. Turn valve to sterilizer.
7. After sterilizer, cycle is finished (timer off). Turn to exhaust cycle.
8. Allow load five (5) minutes to cool.
9. Unload the unit.
10. Turn off the power.

TAB C-70

REFRIGERATOR LOG

A. **PURPOSE:** To provide a chronological record of refrigerator temperatures monitored at the Fleet Hospital.

B. **DEFINITION:** A hardbound log (record book) containing the minimum essential information required to identify temperature fluctuations.

C. **EQUIPMENT, FORMS, AND SUPPLIES REQUIRED:**

Standard record book.

D. **CRITERIA:**

1. Log must be updated frequently to ensure that an accurate and detailed record of temperatures are maintained.

2. At a minimum, it will be updated four times a day, at 6 hour intervals.

E. **STEPS:**

1. The front cover must be marked with the Fleet Hospital Unit Identification Code (UIC), the title "Reefer Log," and the date of initial entry.

2. Each set of facing pages will be divided into vertical columns. Columns will be labeled "DATE," "NAME," "TIME," "LOCATION," "TEMPERATURE," and action required. (See TAB G-29.)

3. The log will be closed at 2400 each day by drawing a double horizontal line beneath the last entry.

4. When the log is full, it will be closed by marking the date of the last entry on the front cover.

F. **RESPONSIBILITY:**

Laboratory Supervisor.

TAB C-71

HAZARDOUS WASTE

A. **PURPOSE:** To provide guidance for the collection, handling and disposal of hospital generated wastes which have contacted living organisms or may otherwise be considered infectious or hazardous.

B. **DEFINITION:**

1. Background: The operation of health care facilities creates waste materials, some of which are hazardous. A subset of hazardous waste is infectious waste; proper handling of infectious waste is mandatory, to prevent spread of infectious diseases. The methods of handling infectious waste, from its generation to its ultimate disposal, must be adhered to strictly by all hands, without exception.

2. Relationship with Host Nations: It is anticipated that the hospital will be operating, in a wartime or conflict mode, on foreign soil. Close liaison with force planners during the pre-deployment planning phase is essential for the hospital command to determine host nation requirements for handling, storage and disposal of infectious hazardous wastes. Whenever possible, agreements and/or contracts with host nations should be secured for the incineration or sanitary burial of wastes in accordance with the host nation's regulations. During peacetime exercises on U.S. soil, adherence to federal, state and local environmental laws and regulations, partially listed in Appendix A, shall be strictly enforced.

3. Categories of Hospital Generated Waste: It must be clearly understood that the field hospital will generate four distinct categories of waste. Each type will require special handling procedures from generation to disposal. These categories are:

(a) Infectious waste - generated in patient contact, laboratory and surgical areas.

(b) Hazardous waste - usually chemical in nature and generated in the Laboratory, X-ray and Public Works department.

(c) Infectious hazardous waste - generated in the laboratory.

(d) Non-infectious waste - generated in all areas of the hospital.

4. Definitions.

(a) Infectious waste is defined as waste originating from the diagnosis and treatment of people. There are five (5) broad categories of infectious waste recognized by the Centers for Disease Control (CDC): microbiological, blood and blood products, pathological, sharps, and isolation waste. Examples of each of these types include, but are not necessarily limited to, the following:

(1) Microbiological - wastes generated in laboratories processing bacterial, fungal, mycobacterial, or viral materials, such as media-containing plates, tubes, or diagnostic strips; swabs; glass slides; pipettes. Live virus vaccines (including smallpox, yellow fever, rubella, measles, mumps, polio, and adenovirus) and any of the associated equipment for their use also fall into this classification.

(2) Blood and blood products - wastes generated in the collection processing, and use of blood and blood products; tubes for diagnostic blood

collection; items and materials contaminated with blood or blood products that are not designed for cleaning, resterilization, and reuse.

(3) Pathological - pathologic specimens, body tissues, contaminated disposable instruments, and laboratory waste generated in the performance of medical treatment procedures and diagnostic laboratory testing.

(4) Sharps - any diagnostic or therapeutic item possessing a surface capable of piercing human skin, not designed for cleaning, resterilization, and reuse. Examples would include needles for injections, preparation of intravenous medicinals, indwelling cannulae, and diagnostic testing (e.g., lumbar puncture, thoracentesis, paracentesis, etc.); scalpels; and other disposable instruments with a surface capable of piercing human skin.

(5) Isolation waste - wastes generated in the therapy of patients on isolation precautions. Examples would include gowns; gloves; masks; head covers; dressings; disposables basins; paper towels used in isolation rooms; and other such items and materials used in the care of isolation patients that are not designed for cleaning, resterilization, and reuse.

(b) Fomites - an object or item that is not of itself harmful, but may harbor pathogenic microorganisms and serve as a vehicle in the transmission of infections. Examples would include but are not limited to bedding, linen, cloth towels and washrags, diagnostic medical instruments (e.g., stethoscopes, sphygmomanometers, thermometers), and personal items (e.g., razors, toothbrushes, toiletries).

(c) Hazardous waste - any wastes, or combination of wastes, which because of its quantity, concentration, physical or chemical properties may pose a substantial present or potential threat to human health or the environment when improperly treated, stored, transported, disposed of or otherwise managed.

(d) Infectious hazardous waste - any combination of materials and agents that meet the definitions described in 2-4.a. and 2-4.c. above. These wastes will typically be generated in the laboratory when organic pathogens are combined with hazardous chemicals or reagents.

(e) Non-infectious waste - waste generated from non-clinical spaces and waste from patients and their related procedures, where no infection or contagious disease exists.

(f) Storage - the holding of infectious hazardous waste for a temporary period, at the end of which the waste is treated, disposed of, or stored elsewhere.

(g) Treatment - any method, technique, or process designed to change the chemical, physical, or biological characteristics of any infectious hazardous waste so as to render such waste nonhazardous, or less hazardous or safer for transportation, storage or disposal.

(h) Autoclave - an apparatus using steam under pressure for sterilizing medical equipment.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:** N/A.

D. **CRITERIA:**

Hazardous waste is properly handled and disposed.

E. **STEPS:**

1. Handling.

(a) Infectious and infectious hazardous waste.

(1) Ward and laboratory personnel shall utilize personal protective clothing and procedures which would normally be practiced in a traditional health care setting for the control of the spread of disease.

(2) Personnel shall wear disposable gloves, gowns, and shoe and hair covers.

(3) Patient contact and laboratory areas will utilize clearly marked, impervious, containers for the disposal of all sharps. When full, the sharps container shall be securely closed with autoclave tape.

(4) Patient areas will utilize clearly marked containers lined with double plastic bags, the outer bag being an orange autoclavable "biological hazard" bag. These containers will be separate from non-infectious "trash" containers. When full, the inner bag will be sealed with autoclave tape. The outer bag will be sealed with filament reinforced tape and autoclave tape.

(b) Hazardous waste.

(1) Protective equipment, as described in DHHS (NIOSH) Publication No. 81-123 (see Appendix A), will be utilized by personnel handling hazardous waste.

(2) All hazardous waste will be containerized. Ideally, in the original container or containers designed for the collection of such wastes such as those provided with automated laboratory equipment.

(3) Containerized and transporting to storage areas will be accomplished by the waste generator (i.e., lab, x-ray, public works, etc.).

2. Transport and storage.

(a) Infectious waste.

(1) Ward personnel will deliver properly sealed sharps containers and doublebagged infectious waste, to the laboratory temporary holding area, on a regularly scheduled basis. Ideally, this area will be one of low traffic and prohibitive to patient care, smoking, eating, and food or medicinal handling.

(2) Ideally, ward personnel will store and transport multiple bags of infectious waste in large, covered containers (i.e., "GI" cans with tight fitting lids). These containers shall be scrubbed with a germicidal solution at least once per shift or more often if grossly contaminated.

(3) Laboratory personnel will handle and routinely autoclave waste under steam pressure for a minimum of fifteen(15) minutes. After proper autoclaving, these wastes may be handled as noninfectious depending on host nation requirements.

(b) Hazardous waste.

(1) As noted in paragraphs 3-1 b.2, hazardous waste will be stored in their original containers or those designed for collection of such wastes.

(2) Waste generating personnel will containerize waste according to its chemical grouping such as lubricants, fuels, acids, alkalines, chlorinated hydrocarbons, etc. Containers will be tightly sealed and labeled.

(3) Storage areas will be at least 100 yards from the hospital

compound and actual or potential potable water sources. Ideally, these areas will be elevated with natural drainage away from the hospital and water sources. Waste containers should be protected from the elements and the area clearly marked as "Hazardous Waste Storage."

3. Disposal.

(a) General. It must be understood that, in an operational situation, the methods of waste disposal range from ideal to undesirable. The following disposal methods are intended to guide the hospital command towards utilization of the best disposal method for any given situation.

(1) Host Nation Agreement - Under the Status of Forces Agreement the cognizant Commander-in-Chief (CINC) will negotiate with the host country for disposal services.

(2) The cognizant CINC will provide disposal services utilizing established logistical support channels within the theater of operations such as the Supply Battalion of the Force Service Support Group, or supply ships.

(b) Methods. In the absence of the preferred, above mentioned disposal methods, the following may be utilized.

(1) Non-hazardous/noninfectious waste (including properly autoclaved infectious waste).

a. Burial in a pit as deep as organic equipment will allow and covered with at least two feet of earth. Burial pits should be at least 100 yards from the hospital compound and potable water sources.

b. Burning by mixing with fuel oil until only ash remains. Ash should then be buried as above. Tactical consideration must be given to open burning as smoke may give away the hospitals location.

(2) Hazardous waste.

a. Laboratory chemical waste which contains infectious, organic matter, is to be treated as hazardous as autoclaving of liquids in closed containers is not authorized.

b. Burial in sealed, marked containers, as deep as organic equipment will permit. Burial sites should be lined with plastic sheeting, covered with at least four feet of earth and conspicuously marked. Sites should be at least 100 yards from the hospital compound and potable water sources.

F. RESPONSIBILITY:

1. The Commanding Officer is responsible for ensuring the proper management of the overall infectious and hazardous waste program and to interface with the host nation to ensure local regulations are satisfied.

2. Nursing Service via the clinical staff is responsible for the handling of all wastes generated in clinical spaces. This includes ensuring that adequate supplies of hampers, bags, tapes, sharps containers, and protective clothing are maintained in these spaces.

3. Laboratory Service is responsible for handling hazardous infectious wastes once it is delivered to or generated by the laboratory. The service is also responsible for proper autoclaving of such wastes to render it free from pathogens.

4. Surgical Service is responsible for handling wastes generated within the operating room giving special attention to surgically removed human tissue.

5. Operating Management is responsible for the removal of waste from the central collection points, including the laboratory, and delivery to the designated pickup area such as the "back loading dock."

6. Public Works Department is responsible for the removal of wastes from the hospital compound and ensuring its proper disposal as outlined in this SOP.

TAB C-72

SHARP ITEM PRECAUTIONS

A. **PURPOSE:** To dispose of used needles and knife blades in a safe manner. To prevent injury and potential risk of contacting hepatitis, syphilis, malaria, aspergillosis, or aids.

B. **DEFINITION:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Needle rack.
2. Perforated stainless steel box.
3. Needle holder.

D. **CRITERIA:**

1. Needles are never discarded loose in trash receptacles.
2. Knife blades are always removed from handles before reprocessing is done.
3. Sharp objects must be enclosed and secured so they cannot perforate the receptacle.

E. **STEPS:**

1. Upon completion of surgical case, the Surgical Tech will:
 - (a) Separate sharp objects from other instruments.
 - (b) Remove knife blades from handles.
 - (1) Point the blade toward table away from self.
 - (2) Remove blades with a needle holder, never use fingers.
 - (3) Place used blades in a non-penetrable box.
 - (c) Place reusable surgical needles, either on needle rack or loose, into a perforated stainless steel box.
 - (d) Dispose of needles in a needle-destruction unit.
2. CSR Decontamination Technician will:
 - (a) Remove any blades/needles from non-operating room departments in the same manner as the Surgical Technician.
 - (b) Run reusable needles, placed in a perforated stainless steel box through the washer-sterilizer.
3. CSR Collection HM will:
 - (a) Collect needle destruction units every watch and empty contents into a firm, self-closing box with padded adhesive tape to secure the opening.

(b) Collect the firm, self-closing boxes located in operating room support space that contain used knife blades.

(c) Take the sealed, labeled contaminated boxes to Environmental Health Department for final disposition.

4. If accidentally puncture/cut finger with contaminated needle/knife blade, do the following:

(a) Notify area supervisor.

(b) Report to Specialty Treatment Area for first aid.

(c) Complete an incident report on NAVMED 6010/14 form.

F. **RESPONSIBILITY:**

1. Lab Technicians.

2. Environmental Health Department.

TAB C-73

PATIENT PROCEDURES FOR HANDLING EXPATRIATED PRISONERS OF WAR

A. **PURPOSE:** To detail patient handling procedures for expatriated prisoners of war within the fleet hospital.

B. **DEFINITION:**

Expatriated prisoners of war (EPW) - those patients who require treatment who are prisoners of U.S. or allied combat forces.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Restraints(theater command military police or hospital issue).
2. Others as specified in admission procedures (all forms will be marked with the words "Prisoner of War" or "EPW").

D. **STEPS:**

1. Upon presentation of EPW to functional area, notify Security Department.
2. Upon admission to Casualty Receiving, Security will be responsible for the following notifications:
 - (a) Theater command military police (MP) headquarters.
 - (b) Executive Officer.
 - (c) Director of Nursing.
 - (d) Director of Administration.
3. Perform essential life saving care.
4. Inform MP that custody of patient will not be assumed by hospital staff and that MP will retain custody of EPW until relieved by appropriate MP headquarters staff or patient is transferred to EPW holding center (external to hospital).
5. After treatment, have corpsman or litter bearer escort MP and EPW to next functional area charge nurse. Admissions packet, correctly annotated will be delivered by hand to charge nurse.
6. During course of treatment, patient will be guarded by MP and/or restrained until treatment is terminated.
7. Movement to another functional area will be reported to Security.
8. EPW's will be fed either on the ward or in the general mess. If allowed to eat in the general mess, EPW's will be accompanied by MP guards.

E. **RESPONSIBILITY:**

CMAA/Security.

TAB C-74

PROCEDURE FOR PICK-UP AND DELIVERY OF HOSPITAL LAUNDRY

A. **PURPOSE:** It will be logistically impossible to pick up and deliver laundry at each individual ward and CSR. Therefore, this procedure establishes central collection points and the methodology for preparing laundry for turn-in.

B. **DEFINITIONS:** N/A.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Canvas laundry bags.
2. Request for clean linen/laundry.

D. **CRITERIA:** N/A.

E. **STEPS:**

1. Designated Laundry Petty Officer will:

(a) Set up laundry bags, tagging one for bed linen, one for clothing (including patient clothing), and one for contaminated laundry.

(b) Daily at 0800, take the soiled laundry to the nearest Clinical Work Space along with a request for the next day's linen/laundry supply.

(c) Distribute cleaned patient clothing.

2. Linen Control Clerks.

(a) Pick-up and receipt for hospital laundry at each Clinical Work Space.

(b) Collect Requests For Clean Linen/Laundry.

(c) Fill requests submitted the previous day and return cleaned patient clothing.

TAB C-75

PROCEDURE FOR HANDLING AND LAUNDERING CONTAMINATED LINENS

A. **PURPOSE:** The Combat Zone Fleet Hospital will generate a significant amount of contaminated linen within the operating rooms and treatment wards. These items will require special handling and laundering to prevent the spread of infection.

B. **DEFINITION:** Contaminated laundry is defined as those items requiring special disinfection and laundering to preclude the spread of infection.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:**

1. Chlorine bleach solution.
2. Latex gloves.

D. **CRITERIA:** N/A.

E. **STEPS:**

1. Hospital ward personnel will bag contaminated laundry separate from regular laundry. Gloves are to be worn when handling contaminated laundry.

2. Contaminated laundry will be receipted by the Linen Control Clerks and delivered to the laundry.

3. At the Laundry all contaminated laundry will be segregated from that requiring only routine processing.

4. Based on the next day's requirements and current inventory the contaminated laundry will be assigned a processing priority.

5. The contaminated laundry will be processed as follows:

(a) Presoak the contaminated laundry for 60 minutes in a chlorine solution of 50 ppm.

(b) Wash the linen in hot water using a normal cycle.

6. Once laundered these items will be placed in inventory for re-issue.

F. **RESPONSIBILITY:**

The Head, Environmental Health Department is responsible for routinely monitoring the handling and laundering of contaminated items to preclude the spread of infections.

CAUTION: Extreme care must be taken to avoid contact with the contaminated laundry to prevent the spread of infection to laundry and other hospital personnel.

TAB C-76

PROCEDURES FOR RELEASE OF MEDICAL INFORMATION

A. **PURPOSE:** To provide procedures of release of medical information within the hospital.

B. **DEFINITION:** Medical Information - Information contained in the health or dental record of individuals who have undergone medical examination or treatment.

C. **EQUIPMENT, SUPPLIES, AND FORMS REQUIRED:** N/A.

D. **STEPS:**

Upon presentation of requests for medical information refer to procedures contained in the following references:

1. Manual of the Medical Department.
2. Freedom of Information Act, BUMEDINST 5720.8.
3. Personal Privacy and Rights of Individuals Regarding Records, SECNAVINST 5211.5.
4. Availability of Navy Records, Policies, SECNAVINST 5720.42.

E. **GENERAL GUIDELINES:**

1. Information contained in health care records of individuals who have undergone medical or dental examination or treatment is personal to the individual and is therefore considered to be of a private and confidential nature. Information from such health care records, the disclosure of which would constitute a clearly unwarranted invasion of personal privacy, should not be made available to anyone except as authorized by the patient or as allowed by the provisions of Manual of the Medical Department and the Privacy Act of 1974 as implemented by SECNAVINST 5211.5 series.

2. Release of information will be coordinated by the Patient Affairs Officer.

3. Personal information of non-medical nature will not be released.

4. Personnel in the patients chain of command may be provided with information required to conduct command business but will be referred to the Patient Affairs Office.

5. Release of information will conform to local command and superior command policy.

6. All Department Heads shall ensure wide dissemination of this information and compliance with procedures outlined herein.

F. **RESPONSIBILITY:**

1. Director of Administration.
2. Patient Affairs Officer.
3. Charge Nurse or Assistant.

TAB D
CLINICAL POLICIES/GUIDELINES INDEX

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TAB D-1

LABORATORY POLICIES

A. The concept of clinical relevance of medical laboratory data takes on new dimensions in the combat zone. The concept must be influenced by requirements for mobility of both the patient and the medical treatment facility. Both are expected to be moving faster and more frequently in future conflicts.

B. Arterial blood gas determination is desirable for Echelon 3 and 4; however, there is not presently a liquid blood gas machine that will meet the Reliability Availability Maintainability characteristics. This requirement is essential, but not critical to delivery of health care in field hospitals.

C. Culture and sensitivity testing will be accomplished only at Echelon 4.

D. Specimens from patients presenting conditions of significant epidemiological concern that cannot be analyzed in Echelon 3 or 4 laboratories will be referred to the Theater Area Laboratory and/or CONUS reference laboratory for evaluation. Specimens will be refrigerated (if applicable) and placed in the sample chain as soon as possible.

E. Echelon 3 facilities deployed to Southeast Asia, Northeast Asia, Southwest Asia, Central America, Middle East, or Africa's theater of operation will request the Microbiology Augmentation Set to supplement laboratory capabilities.

F. Blood Bank Policies:

1. Deployable Medical Systems (DEPMEDS) will use the 2004 blood study as a basis for blood bank operation.

2. There will be four units of blood per wounded in action (admitted to Echelons 3 and 4 facilities).

3. Both fresh and frozen red cells will be available in the theater after D plus 15. During the first 15 days, frozen red blood cells will be used.

TAB D-2

LABORATORY GUIDELINES

A. Procedures performed at NATO Echelons 3 and 4 are as follows:

1. Chemistry:

Task No.	Description	Echelon 3	Echelon 4
E002	Determine electrolyte levels (NA, K, CL, C02)	X	X
E003	Determine total serum protein level	X	X
E005	Determine serum creatinine level	X	X
E007	Determine serum amylase level	X	X
E008	Determine SGPT level	X	X
E009	Determine CPK level	X	X
E010	Determine blood glucose level	X	X
E011	Determine BUN level	X	X
E012	Determine serum bilirubin level	X	X
E013	Determine spinal fluid sugar level	X	X
E014	Determine spinal fluid protein level	X	X
E017	Determine calcium level	X	X
E015	Determine SGOT level		X

2. Hematology/Urinalysis:

E020	Perform complete blood count (WBC,HGB,HCT)	X	X
E021	Perform white cell count	X	X
E022	Determine hematocrit level	X	X
E024	Perform white cell differential count	X	X
E025	Perform prothrombin time (PT)	X	X
E026	Perform partial thromboplastin time (PTT)	X	X
E028	Perform spinal fluid cell count & Differential	X	X
E029	Perform urinalysis w/specific gravity	X	X
E030	Perform microscopic urinalysis	X	X
E031	Perform platelet estimate	X	X
E032	Perform platelet count	X	X
E033	Determine fibrinogen level & fibrin split products		X
E027	Perform occult blood determination	X	X
E043	Perform gram stain	X	X
E044	Perform RPR test for syphilis	X	X
E045	Perform mononucleosis spot test	X	X
E046	Perform thick and thin smear for malaria	X	X
E047	Examine feces for ova, cysts & parasites	X	X
E048	Perform potassium hydroxide (KOH) Preparation	X	X
E049	Perform pregnancy determination	X	X

4. Blood Bank:

E052	Perform blood T&C (ABO, RH) (2 units)	X	X
E057	Perform thaw & wash of frozen RBCs (2 units)	X	X
E058	Perform thaw & wash of frozen platelets (6 pack)	X	X
E059	Perform thaw of frozen plasma	X	X

TAB E
STANDARDS AND JOB DESCRIPTIONS INDEX

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TAB E-1

LABORATORY DEPARTMENT CLEANING SCHEDULE

A. **PURPOSE:** To maintain the cleanliness of the department and keep all areas free from any environmental hazard.

B. **EQUIPMENT AND SUPPLIES NEEDED:**

1. 1 Mop bucket and mop squeeze.
2. 2 Mop handles and removable mop heads.
3. 2 Push brooms.
4. 1 Disinfectant solution.
5. 4 Sponges.
6. 1 Dust pan.
7. 1 Foxtail.
8. Laundry bags.
9. Plastic trash bags (small and large liners).

C. **CRITERIA:**

1. Damp dust all tables, shelves, and equipments before the end of each shift.
2. Daily cleaning schedule will not exceed 30 minutes.
3. Trash and all medical waste are properly disposed after each procedure.
4. Specific working areas will be cleaned and squared away at the end of each shift.
5. Temper tents are cleaned every Thursday. Change ISO container vent covers weekly.

D. **DAILY CLEANING SCHEDULE:**

1. Damp dust specific assigned spaces before the end of each shift.
2. Wash sinks in each assigned space at the end of each shift.
3. Wipe counters and table tops with disinfectant solution (NSN: 6840-01-066-7466, PG).
4. Allow all surfaces to air dry.
5. Organize all equipments and restock supplies as needed.

E. **WEEKLY CLEANING SCHEDULE:**

1. Wash down ISO container bulkheads with disinfectant (NSN: 6840-01-066-7455, PG) solution every Thursday - to be done by day shift.
2. Change vent covers to ISO containers - to be done by day shift.

3. Launder cotton liners of Temper tent at discretion of supervisor - to be prepared and reassembled by night shift.

4. Log cleaning in the Laboratory Department Daily Log maintained by the leading Chief Petty Officer.

F. **RESPONSIBILITY:**

Laboratory Technicians.

TAB E-2

PERSONAL STANDARDS FOR LABORATORY PERSONNEL

1. The prescribed uniform of the day will be worn at all times or as directed.
2. Lab coats or smock jackets can be worn in the department as directed.
3. Eating and drinking are prohibited in laboratory spaces.
4. All laboratory specimens are to be treated as though they are infectious. Personnel must wipe up spills immediately with disinfectant solution (FSN: 6840-01-006-7466, PG) and must wash hands frequently.

TAB E-3

HEAD, LABORATORY DEPARTMENT JOB DESCRIPTION

A. **RESPONSIBILITIES:** Responsible and accountable for the management of all functions and services performed by the Laboratory Department.

SPECIFICALLY HE WILL:

1. Set policies and procedures for running the Laboratory Department.
2. Approve all communications within and outside of the Department.
3. Approve all personnel performance evaluations.
4. Prepare and submit all Departmental reports in final form.
5. Perform autopsies.
6. Provide advice and consultation concerning clinical and anatomic pathology.

B. **QUALIFICATIONS:**

1. Designator 2100/0-5
2. Billet number 70020.00
3. Licensed Physician: Board Eligible or Board Certified Clinical and Anatomic Pathologist.
4. ACLS certified.

TAB E-4

LABORATORY OFFICER JOB DESCRIPTION

A. RESPONSIBILITIES:

1. Supports the Head, Laboratory Department by implementing lab policies.
2. Supervises the operation and maintenance of the laboratory.
3. Monitors all quality control procedures and results.
4. Performs administrative functions related to laboratory personnel, equipment, and supplies.
5. Oversees an inservice education and training program.

B. QUALIFICATIONS:

1. Designator 2300/0-4; NOBC 0866.
2. Billet number 72020.
3. BCLS certified.

TAB E-5

LEADING CHIEF PETTY OFFICER JOB DESCRIPTION

A. RESPONSIBILITIES:

1. Direct, assist, orient, and instruct staff in principles, procedures, and safety precautions employed in Laboratory Department.

2. Supervise, schedule, and coordinate activities of departmental personnel with Module LPO's.

3. Supervise supplies procurement and make recommendations for improvement, replacement, or purchase of products.

(a) Prepare supply requisitions.

(b) Maintain supply storerooms, and monitor stock level.

4. Interpret and implement hospital policies and procedures applicable to Laboratory Department.

5. Evaluate the performance of all enlisted personnel.

(a) Maintain anecdotal notes on personnel.

(b) Prepare enlisted performance evaluations as required.

6. Coordinate leave/liberty for enlisted personnel. Make staff assignment changes as necessary.

7. Ensure that all hands are knowledgeable of all safety related codes (i.e., fire and evacuation flow charts) during both drills and actual emergencies.

8. Prepare and submit changes to the Laboratory Department Procedure Manual for approval by the Head, Laboratory Department.

9. Develop and prepare departmental report.

10. Prepare monthly morbidity report.

11. Muster personnel and inform them of the plan of the day and any changes.

12. Oversee the accessioning area of the laboratory.

13. Prepare work requests and monitor progress.

14. Prepare and submit Fire and Watch Bills monthly.

15. Assist the Laboratory Officer and Head, Laboratory Department.

16. Identify educational needs for the department and help plan and teach the programs.

B. QUALIFICATIONS:

1. NEC 8506/C-7.

2. Billet Number 72010.

3. BCLS certified.

TAB E-6

PM SHIFT LPO JOB DESCRIPTION

A. RESPONSIBILITIES:

1. Supervise, schedule, and coordinate activities of the PM shift personnel with the Module LPO's.
2. Muster personnel and inform them of the plan of the day and any changes.
3. Direct, assist, orient, and instruct staff in principles, procedures, and safety precautions employed in laboratory department.
4. Oversee the accessioning area of the laboratory.
5. Ensure that all hands are knowledgeable of all safety related codes (i.e., fire and evacuation flow charts) during both drills and actual emergencies.
6. Assist the Laboratory Officer and Head, Laboratory Department.
7. Perform duties as assigned by LCPO regarding the PM shift.

B. QUALIFICATIONS:

1. NEC 8506/E-6.
2. Billet number 72030.00.
3. BCLS certified.

TAB E-7

MODULE LPO JOB DESCRIPTION

A. RESPONSIBILITIES:

1. Make daily personnel assignments within his module. Set the work pace and priority. Prepare daily schedule.
2. Supervise performance of subordinates, including that of professional and military nature.
3. Ensure that all tasks are properly completed and safety standards are met.
4. Maintain high standards of personal hygiene and conduct.
5. Maintain clean spaces.
6. Check and maintain daily availability of equipment/supplies and submit order requests to the Watch LPO.
7. Ensure that day logs and daily record sheets have been completed correctly.
8. Report to and obtain assistance from the Watch LPO as needed.
9. Pass word to oncoming watch.
10. Review all lab results within his module.
11. Complete the daily morbidity statistics each working day.
12. Review results of all equipment maintenance and quality control procedures.
 - (a) Advise the Watch LPO on all matters of equipment status.
 - (b) Advise the Laboratory Officer on all quality control problems.
13. Perform other duties as assigned.
14. Advise the Watch LPO of necessary changes to procedures.
15. In the absence of the Watch LPO, may be assigned in his place.

B. QUALIFICATIONS:

1. NEC 8506/E-6.
2. Billet numbers 72030.01-04.
3. BCLS certified.

TAB E-8

ADVANCED LABORATORY TECHNICIAN JOB DESCRIPTION

A. RESPONSIBILITIES:

1. Perform advanced laboratory procedures in all sections of the laboratory including Blood Bank, hematology, chemistry, urinalysis, microbiology, and serology.
2. Perform blood gas determinations.
3. Perform quality control procedures, equipment preventive maintenance, and basic equipment troubleshooting.
4. Prepare and submit reports and maintain records as required.
5. Supervise, instruct and evaluate junior personnel.
6. Perform other duties as assigned.

B. QUALIFICATIONS:

1. NEC 8506/E-6 - E-4.
2. Billets numbers 72030.05, 72050.00 - 09, 72070.00 - 03.
3. BCLS certified.

TAB E-9

BASIC LABORATORY TECHNICIAN JOB DESCRIPTIONS

A. RESPONSIBILITIES:

1. Perform basic laboratory procedures in the hematology, chemistry, urinalysis, microbiology, and serology sections of the laboratory.
2. Perform venipuncture.
3. Perform preliminary screening of blood donors and donor phlebotomy.
4. Perform quality control procedures.
5. Maintain records as required.
6. Perform other duties as assigned.

B. QUALIFICATIONS:

1. NEC 8501/ E-3.
2. Billet number 76090.00 - 02.
3. BCLS certified.

TAB F
REFERENCES INDEX

<u>NUMBER</u>	<u>TITLE</u>
F-1	"Textbook of Clinical Chemistry," Ed Norbert, W. Tiets, Saunders Co. Philadelphia
F-2	"AABB Technical Manual," Ninth Ed., Arlington, VA Ed. Frances K. Widmann, M.D.
F-3	"Clinical Diagnosis and Management by Laborator Methods" Volumes I & II. Ed. John Bernard Henry, M.D., W. B. Saunders Co., Philadelphia.
F-4	"Urinary Sediment, A Textbook Atlas," Meryl H. Haber, ASCP Educational Products Division.
F-5	"Manual of Clinical Microbiology," 2ND Ed., Ed. Edwin H. Lennette, ASM, Washington, D.C..
F-6	NAVMED P-5123, Operational Procedures for Military Blood Donor Centers, Armed Services Whole Blood Processing Laboratories, and Blood Transshipment.
F-7	"The Morphology of Human Blood Cells," L. W. Diggs, Dorothy Storm, Ann Bell, Abbott Laboratories, North Chicago, IL 60064.
F-8	"Use of the Gram Stain in Clinical Infections Diseases," Schering Corporation, Kenilworth, N.J. 07033.
F-9	NAVMED P-5083, Methods for Preparing Pathology Specimens for Storage and Shipping.
F-10	NAVMED P-5120, Standards of American Association of Blood Banks.

TAB G
FORMS INDEX

<u>NUMBER</u>	<u>FORM NUMBER</u>	<u>FORM TITLE</u>	<u>PAGE</u>
G-1	SF 546	Chemistry I	
G-2	SF 547	Chemistry II	
G-3	SF 548	Chemistry III (Urine)	
G-4	SF 549	Hematology	
G-5	SF 550	Urinalysis	
G-6	SF 551	Serology	
G-7	SF 552	Parasitology	
G-8	SF 553	Microbiology I	
G-9	SF 554	Microbiology II	
G-10	SF 555	Spinal Fluid	
G-11	SF 556	Immunohematology	
G-12	SF 557	Miscellaneous	
G-13	DD 572	Blood Donor Record	
G-14	SF 518	Blood or Blood Transfusion	
G-15	DD 573	Shipping Inventory of Blood Products	
G-16	NAVMED 6530/1	Blood Bank Operational Report	
G-17	SF 541	Gynecological Cytology	
G-18	SF 542	Specimen Record	
G-19	SF 515	Tissue Exam	
G-20	NA	Hematology Log Format	210
G-21	NA	Urinalysis Log Format	212
G-22	NA	Chemistry Log Format	214
G-23	NA	Mail Out Log Format	216
G-24	NA	Blood Bank Crossmatch Log Format	218
G-25	NA	Blood Drawing Log Format	220
G-26	NA	Exit Questionnaire	221

G-27	NA	Reefer Temperature Log	222
G-28	DD-599	Patient's Effects Storage Tag	
G-29	NAVMED 6010/8	Patient's Valuables Envelope	

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BLOOD BANK CROSSMATCH LOG

RECIPIENT TRANSFUSION NUMBER	RECIPIENT NAME	TECH REGISTRATION NUMBER	RECEPIENT ABO/RH	T&C RESULT	DOING TESTING
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BLOOD BANK CROSSMATCH LOG

DONOR UNIT NUMBER	DONOR ABO/RH	UNIT EXPIRATION DATE	ISSUED BY	DATE & TIME ISSUED	ISSUED TO	DISPOSITION
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[illegible]

BLOOD DRAWING LOG

TAB G-26

EXIT QUESTIONNAIRE

DONOR
NUMBER _____

I wish my blood donation to be used for:

_____ Transfusion to patients.

_____ Research ONLY. My blood may not be suitable for transfusion
to patients.

MARK ONLY ONE ANSWER.

PLEASE COMPLETE AND PLACE IN COLLECTION BOX.

REFRIGERATOR TEMPERATURE LOG FORMAT

DATE	TIME	LOCATION	TEMPERATURE
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TAB G-27

REFRIGERATOR TEMPERATURE LOG FORMAT

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